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Growth Through Agricultural Progress

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Paul R. Miller

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CONTENTS

PRELIMINARY STUDIES ON CONTROL OF SOUTHWESTERN COTTON RUST[1]

Lester M. Blank[2]

Abstract

Fungicides for control of cotton rust, Puccinia stakmanii, have been tested in greenhouse and field experiments. The results indicate that zineb (65% W.P.) at 2 pounds per 40 gallons of water per acre is very effective when applied as a foliage spray prior to infection of the cotton plant by the rust fungus.

Southwestern rust of cotton, caused by Puccinia stakmanii Presley (= Puccinia cacabata Arth. & Holw.), has been known for many years in Texas, New Mexico, and Arizona and in northern Mexico on Gossypium hirsutum and G. barbadense. The aecial stage on cotton was reported first in 1897 (6) and its association with the uredial and telial stages on grama grass (Bouteloua spp.) was established in 1942 (7). In Arizona the overwintering teliospores on the grass are activated by the summer rains in July, and sporidia are produced and released to drift to nearby cotton fields. Within 12 to 15 days after infection the orange-yellow aecia are evident on the under side of cotton leaves, on bracts, and on young bolls. Shedding of leaves, squares, and young bolls follows within several weeks and may result in extensive disease losses (3, 4, 8, 9, 10). In a replicated test in Sonora, Mexico, Duffield (5) established various degrees of intensity of infection on field-grown plants by exposing them for different periods of time to sporidia from infected grama grass. The reduction in yield of seed cotton from plots of "severe" rust was 39%, from "moderate" rust 21%, and from "light" rust less than 1%. The fluctuating intensity of and damage by rust infection from locality to locality and from year to year are typical of rust. The importance of the amount of potential inoculum carry-over and of the amount and frequency of summer rains in relation to subsequent development of rust in epiphytotic proportions has been noted (9).

Efforts to control rust have been limited and generally unproductive. A localized program of eradication of grama grass in and around cotton fields was suggested as a beneficial measure (8), but the probability exists that spores may be carried for miles on currents of air (3, 4). Applications of fungicides on the cotton plant have been reported intermittently in Arizona (1, 2, 4), but in none of the tests was development of rust sufficient to warrant conclusions. Duffield (5), in Mexico, applied four fungicides claimed to have eradicative activity 8 and 16 days after the field plants had been exposed to rust inoculum. No control was demonstrated, and none of the materials had any visible effect upon the subsequent development of the rust fungus.

A number of commercial varieties of Upland (Gossypium hirsutum) and American-Egyptian cotton (G. barbadense), as well as wild and Old World cultivated species of Gossypium, have been tested for reaction to rust. All the commercial varieties grown in this hemisphere were highly susceptible. Several of the other species were reported to be mildly susceptible or resistant (5, 8).

EXPERIMENTAL RESULTS

Experimental work on the use of fungicides for control of cotton rust was started in 1956 in the greenhouse and the field. In the greenhouse tests, naturally infected grama grass was soaked for an hour or more before it was placed on a screen in a high-humidity chamber. Usually within 12 to 24 hours, at about 70° to 85° F, production of sporidia began and continued for several days. Seedling cotton plants were exposed to the rust inoculum for 12 to 24 hours and incubated overnight in another humidity chamber before being moved to the open greenhouse. Lesions were evident within 5 to 8 days, and notes on the number of lesions were made about 10 to 12 days after inoculation. Fungicides were applied as sprays to the point of runoff, before or after inoculation of the plants. The tests demonstrated that many of the fungicides were ineffective in controlling rust, that others were partially, and that some were very effective. Best control was obtained when the sprays were applied shortly before exposure of the plants to the rust inoculum, and none was effective when applied after such exposure.

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Field testing of fungicides was done in plots located in cotton-production areas of southern Arizona where rust had been prevalent in previous years. Rust development was inadequate in 1956 and 1957 to permit evaluation of test materials, but in 1958 satisfactory disease development occurred on three of the six plot locations. Sixteen fungicides were evaluated in non-replicated plots at each of these locations, and three were considered highly effective. They were zinc ethylene bisdithiocarbamate (zineb), n-dodecylguanidine acetate (dodine), and an experimental material composed of zineb and nickel chloride.

In 1959 field testing was done at four locations, using the three most promising materials of 1958 along with four additional fungicides. Treatments were made in duplicate at each location on July 8, 22, and 30. Rust was severe at the three locations in Cochise County, with the heaviest fall of sporidia occurring about the time of the last application of the fungicides. Again the most effective fungicides appeared to be zineb, dodine, and the zineb-nickel chloride mixture. Plots receiving these fungicides had fewer rust lesions than the controls and subsequently showed little or no defoliation.

The 1960 tests were conducted on three farms in Cochise County, one near Kansas Settlement community and two north of Elfrida. The individual treatments were applied to plots four rows wide and 25 feet long, with three replications in randomized block design. The fungicides plus sticker-spreader additive were applied with a knapsack sprayer, and good coverage of the plants was obtained with the solutions applied at the rate of 40 gallons/acre. The treatments were applied June 29 and July 11 on these plots, and on July 21 and August 1 new footages of row were sprayed without replication. The heaviest infection period occurred in the latter part of July, and examination of all plots on August 10 revealed a high degree of control with zineb (65% W.P. at 2 pounds/acre). Somewhat less control occurred with dodine and with zinc dimethyldithiocarbamate (ziram). Tetramethylthiuram disulfide (thiram) was ineffective, and these plots appeared to have as many lesions and as severe defoliation as the untreated controls. The plants on the zineb plots, which had fewer lesions than the controls, retained their foliage and continued active growth and fruiting processes.

Yield data were obtained from the first harvest of two of the replicated tests, and second and final harvest data were available in but one of these tests. Considering only the data from first picking, at which time an estimated 90% of the total yield was obtained, the zineb-treated plots invariably surpassed the untreated plots of the same replicate in yield of seed cotton. The yield associated with zineb treatment was about 50% above that of the untreated controls. The overall performance of the other fungicides suggested that slight increases in yield were obtained with dodine and ziram, whereas the thiram treatment was without appreciable effect. Because of the limited number of tests and the lack of complete yield data, no attempt is made to evaluate critically the effect of fungicides on yield.

In addition to the replicated tests discussed, observation was made of large-scale applications of zineb fungicide for control of rust. Prior to the 1960 crop season, an informal presentation of findings of 1958 and 1959 in regard to control measures was made to the growers of the area. As a result, a number of growers applied zineb to their fields on a commercial basis. Where treatments were made prior to development of rust epidemics, the results appeared to be very similar to those observed and recorded in the experimental plots. Applications made after the period of heaviest infection were of no benefit.

These preliminary studies on control of cotton rust with fungicides indicate that zineb is very effective when applied as a foliage spray prior to infection of the cotton plant by the rust fungus.

Literature Cited

1. ANONYMOUS. 1943. Rust of cotton. Arizona Agr. Exp. Sta. Ann. Rept. 75-76.
2. BERKENKAMP, B. B. 1958. Studies on southwestern cotton rust (Puccinia cacabata Arth. & Holw.). MS Thesis, University of Arizona.
3. BROWN, J. G. 1938. Cotton rust in Arizona. Plant Disease Reptr. 22: 380-382.
4. BROWN, J. G., and R. B. STREETS. 1934. Diseases of field crops in Arizona. Arizona Agr. Exp. Sta. Bull. 148.
5. DUFFIELD, P. C. 1958. Biology of Puccinia stakmanii. PhD. Thesis, Iowa State College, Ames, Iowa.

6. ELLIS, J. B., and B. M. EVERHART. 1897. New West American fungi. III. Erythea 5: 5-7.
7. PRESLEY, JOHN T. 1942. Aecidium gossypii, the aecial stage of Puccinia boutelouae. Phytopathology 32: 97-99.
8. PRESLEY, JOHN T., and C. J. KING. 1943. A description of the fungus causing cotton rust, and a preliminary survey of its hosts. Phytopathology 33: 382-389.
9. SMITH, T. E. 1960. Observations on cotton rust (Puccinia stakmanii) under severe disease conditions. Plant Disease Reptr. 44: 77-79.
10. TAUBENHAUS, J. J. 1917. On a sudden outbreak of cotton rust in Texas. Science (n. s.) 46: 267-269.

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PHLEOMYCIN, AN ANTIBIOTIC MARKEDLY EFFECTIVE FOR CONTROL OF BEAN RUST

B. C. Smale, M. D. Montgillion,[1] and T. G. Pridham[2]

Summary

The antibiotic phleomycin was shown in greenhouse studies to be a markedly effective therapeutant and protectant for bean rust (Uromyces phaseoli (Pers.) Wint. var. typica Arth.). The effects of phleomycin at a concentration of 5 ppm applied to upper surfaces of primary leaves at intervals of 5, 2, or 1 day before or after inoculation of their lower surfaces indicated that the absorbed antibiotic exerted a high degree of both protectant and curative action against the rust fungus. The ED-50 value for phleomycin applied to the upper primary leaf surfaces 1 hour before inoculation of the lower leaf surfaces was 0.1 ppm. Inoculation of the lower leaf surfaces, which 1 hour earlier had been sprayed with phleomycin, resulted in an ED-50 value of only 0.01 ppm. No disease occurred when primary leaves were inoculated within 24 hours after application of low concentrations of the antibiotic to roots or stems. Data from studies involving application of the antibiotic to proximal or distal half-leaves suggested that translocation of the antibiotic is associated with the transpiration stream.

INTRODUCTION

Phleomycin[3] was described first by Maeda, et al. (2) and subsequently studied in more detail by Takita (8) and Takita, et al. (8, 9). The antibiotic is produced by Streptomyces verticillus Okami, Suzuki, & Umezawa (strain 843-1). Streptomyces verticillus also produces a heptaene antifungal antibiotic and at least two other antibacterial antibiotics. The exact chemical nature of phleomycin is not clear. Japanese workers initially stated that it contained carbohydrate and peptide moieties and proposed an empirical formula of $C_{13-14}H_{28-29}N_3O_{12}$. Later, the empirical formula was revised to $C_{53}H_{91}N_{17}O_{32}Cu$ or $C_{53}H_{93}N_{17}O_{32}$. Their reports suggest some question as to whether copper is a component of the molecule as synthesized by the organism. Copper-free preparations exhibit about the same degree of in vitro antibiotic activity as do those containing the element. The antibiotic apparently is basic and water soluble.

Our attention was directed to phleomycin in connection with efforts to determine the nature of the anti-bean rust agent F-17 produced by Streptomyces cinnamomeus f. azacoluta Pridham, et al. (strain NRRL B-1699) (3, 4, 5). Mitchell, et al. (3) have shown that some agent present in culture filtrates obtained with this strain possesses marked therapeutic and protectant properties for bean rust. The name of the Japanese culture suggests that S. verticillus and S. cinnamomeus f. azacoluta are closely related. Moreover, the qualitative patterns of antibiotic production and the methods for isolation of the several factors produced by both strains are similar. It has been established that duramycin, one of the antibacterial antibiotics, and F-17-C, the antifungal heptaene complex produced by S. cinnamomeus f. azacoluta, are not responsible by themselves for the anti-bean rust activity of culture filtrates (7).

This report concerns phleomycin as a potential plant therapeutant and protectant for bean rust and contains information on the absorption, translocation, and efficacy of the antibiotic when applied to bean plants.

METHODS

Preparation of Phleomycin Solutions: Threefold serial dilutions of phleomycin[4] with associated copper and of the same degree of purity as that described by Takita (8) were prepared with distilled water containing 0.1% Tween 20. Concentrations of 42 through 0.006 ppm were used in leaf and stem applications. Root applications were made with threefold serial dilutions

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[3]Phleomycin was supplied by Bristol Laboratories, Syracuse 1, New York.

[4]The phleomycin preparation used here is viewed as composed of a single antibiotic, but it should be emphasized that this preparation may contain more than one antibiotic factor.

in tap water (no Tween 20) ranging in concentration from 1.6 through 0.02 ppm.

Test Plants: Pinto bean plants were selected for uniformity when the primary leaves were fully expanded and the trifoliolate leaves were tightly folded within the terminal bud. Experiments involved five plants per treatment. These experiments were repeated two or three times except in the root-absorption studies where data were obtained from a single experiment with four plants per concentration.

Leaf Application: In experiments involving therapy or translocation, antibiotic solutions were applied to the entire upper surface of primary leaves or portions of them, with cotton swabs saturated with phleomycin solutions. One-tenth to two-tenths ml was required to treat the entire upper surface of a primary leaf uniformly.

Protectant properties of the phleomycin residual on the surface of leaves were studied using a method previously described (6). This method involved spray inoculation of the air-dried lower surfaces of primary leaves previously sprayed to run-off with aqueous solutions of the compound. The quantity of inoculation-spray was limited so that redistribution of the dry phleomycin coating on the leaves would be minimized. Primary leaves of plants used as controls were sprayed similarly with the aqueous carrier, allowed to dry, and spray-inoculated in the same manner.

Stem Application: Phleomycin was applied to stems by rolling a cotton swab saturated with a phleomycin solution over the entire surface of hypocotyl and first internode.

Root Application: Root-absorption studies were carried out with tap water solutions of phleomycin, using the plant liquid-culture apparatus of Linder and Mitchell (1). Two plants with soil removed from the roots were placed in tap water in each of the culture vessels. Plants were kept for 4 hours at temperatures of about 20°C and were illuminated by fluorescent tubes which produced about 800-foot-candle intensity. After this period of adjustment, the tap water was removed from the culture vessels and replaced with fresh tap water containing phleomycin. Lower surfaces of primary leaves were inoculated 24 hours later. Four plants were employed for each concentration of phleomycin and for the controls.

Inoculation: A suspension of Uromyces phaseoli (Pers.) Wint. var. typica Arth. uredospores (0.5 mg per ml of water) was applied uniformly to the under surface of primary leaves with an artist's water-color brush. Uredospores used for inoculum were collected from diseased plants maintained in the greenhouse about 3 weeks. A suitable water suspension of uredospores was obtained by first mixing spores with a small amount of Tween 20 and then adding distilled water. The amount of Tween used resulted in a 0.1% concentration of Tween. A homogeneous suspension of spores was maintained throughout brush-inoculation procedures by use of a magnetic stirrer.

Incubation: After inoculation, or treatment and inoculation, the plants were immediately placed in a clear plastic-covered enclosure and mechanically humidified for about 15 seconds every 10 minutes. During the incubation period of 24 hours, plants were supplied with 12 hours of fluorescent illumination (800-foot-candle intensity) and were kept at a temperature of about 20°C. Plants were subsequently moved into the greenhouse except in root application studies where plants were kept at temperatures of about 20° and illuminated daily for 12 hours by fluorescent tubes which produced a 3000-foot-candle intensity.

Ten days after inoculation, disease severity in the untreated inoculated controls and in the various treatments was estimated on a scale of 0 through 10 (10 representing maximum disease), and percentage control of rust computed.

RESULTS AND DISCUSSION

Application to Leaves Immediately Before Inoculation: Complete inhibition of bean rust symptoms was obtained when the upper primary leaf surfaces were treated with 42, 14, 4.7, or 1.6 ppm solutions of phleomycin immediately before inoculation of the lower primary leaf surfaces (Fig. 1). The marked efficacy of the absorbed antibiotic under these conditions is indicated by the low ED-95 and ED-50 values of 0.5 and 0.1 ppm, respectively. On the basis of volume of solution applied (0.2 ml/leaf), only 0.1 microgram of phleomycin per leaf was required to produce the low ED-95 value even though the antibiotic was applied to the opposite surface from the one inoculated. Such fungitoxicity was in marked contrast to phytotoxicity, which was not apparent when 30 times this amount was applied and was only moderate when 90 times this amount was applied.

The marked protectant properties of phleomycin were demonstrated by spray inoculation of lower leaf surfaces treated 1 hour earlier with the antibiotic (Fig. 1). The ED-50 of phleomycin under such conditions was 0.01 ppm.

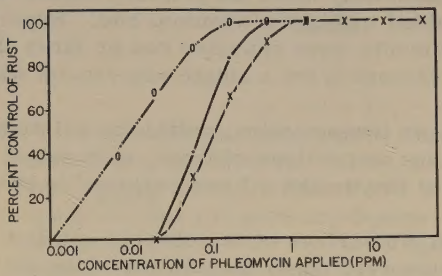

FIGURE 1. Phleomycin-Dosage-Response Curves with Bean Rust. The antibiotic in aqueous solution was applied to roots 24 hours before or to leaves 1 hour before inoculation. Solid-line curve - phleomycin applied to roots; 0-point curve - phleomycin applied to the leaf surfaces that were subsequently inoculated (that is, lower surface); X-point curve - phleomycin applied to the opposite leaf surfaces (that is, upper surfaces) from those subsequently inoculated.

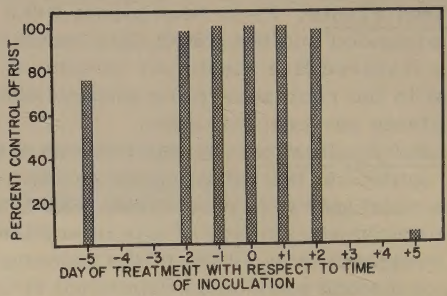

FIGURE 2. Protective and Therapeutic Effects of Absorbed Phleomycin on Bean Rust. Bars indicate disease control obtained when approximately 0.2-ml portions of a 5 ppm aqueous solution of phleomycin were applied to the upper surfaces of the primary leaves of beans at the designated intervals before, at the time of, or after inoculation of the lower surfaces of the primary leaves.

Application to Leaves at Intervals Before or After Inoculation: Application of a concentratration of 5 ppm of phleomycin to upper surfaces of primary leaves at intervals of 5, 2, or 1 day before or after inoculation of the lower surfaces of the primary leaves indicated that the absorbed and translocated antibiotic exerts both protective and curative action against the rust (Fig. 2). Complete or nearly complete control of rust was obtained with phleomycin for the period extending from 2 days before through 2 days after inoculation. The much greater efficacy of the absorbed antibiotic applied 5 days before inoculation than that applied 5 days after inoculation was not surprising since the first symptoms of rust appear 6 days after inoculation. It is interesting that the concentration of antirust activity within the leaf 5 days after application of only 1 microgram of phleomycin to the leaf surface was sufficiently high to reduce rust symptoms by about 75%.

Application to Proximal and Distal Portions of Leaves: Translocation of phleomycin in leaves, like that of most other antibiotics, is associated with the transpiration stream. Application of a 14 ppm concentration of the antibiotic to the distal half of primary leaves followed by inoculation of the entire lower surface of these leaves prevented pustule formation in the distal portions, but no reduction in pustule number occurred in the proximal portions of leaves. As would be expected, treatment of proximal portions of leaves followed by inoculation of the lower surfaces prevented development of rust pustules over the entire leaf area.

Application to Stems: Application of a 14 ppm concentration of phleomycin to stems immediately before inoculation of primary leaves resulted in absorption of the antibiotic by the stems and translocation of the fungitoxic compound to primary leaves (5 to 8 cm distant) in sufficient amounts to prevent development of rust completely.

Application to Roots: Phleomycin was absorbed by roots and readily translocated to primary leaves where it accumulated in disease-controlling amounts. Absorption of phleomycin from aerated tap water containing concentrations of 1.6 and 0.56 ppm (160 and 56 micrograms total antibiotic available per two plants) and subsequent translocation to leaves resulted in complete disease control (Fig. 1). These data on root absorption are preliminary and, although interesting in comparison with the efficacy of leaf-applied antibiotic, should be considered as suggestive only.

This investigation demonstrates the unusual nature of phleomycin in that exceptionally low concentrations of the antibiotic are required for the control of bean rust and that no phytotoxic effects are noted at concentrations several hundredfold in excess of disease-controlling levels. Although experiments on the period of effectiveness of absorbed and translocated phleomycin were based on application of a concentration of only 5 ppm, the data suggest that protection from rust by absorbed antibiotic for intervals in excess of the 5 days observed should result if the concentration applied were increased several fold. The role of the copper associated

with the phleomycin in the fungitoxicity observed has not been determined. Takita (8) has shown that removal of the metal did not alter the bacteriostatic activity of phleomycin.

Literature Cited

1. LINDER, P. J., and J. W. MITCHELL. 1960. Rapid transport of α-methoxyphenylacetic acid introduced directly into the water stream of bean plants. Botan. Gaz. 121: 139-142.
2. MAEDA, K., H. KOSAKA, K. YAGISHITA, and H. UMEZAWA. 1956. A new antibiotic, phleomycin. J. of Antibiotics, Ser. A. 9: 82.
3. MITCHELL, J. W., B. C. SMALE, E. J. DALY, W. H. PRESTON, Jr., T. G. PRIDHAM, and E. S. SHARPE. 1959. Absorption and translocation of the F-17 antirust complex by bean plants and subsequent effect on the rust fungus, Uromyces phaseoli typica. Plant Disease Reptr. 43: 431-436.
4. PRIDHAM, T. G., L. A. LINDENFELSER, O. L. SHOTWELL, F. H. STODOLA, R. G. BENEDICT, C. FOLEY, R. W. JACKSON, W. J. ZAUMEYER, W. H. PRESTON, Jr., and J. W. MITCHELL. 1956. Antibiotics against plant disease. I. Laboratory and greenhouse survey. Phytopathology 46: 568-575.
5. PRIDHAM, T. G., O. L. SHOTWELL, F. H. STODOLA, L. A. LINDENFELSER, R. G. BENEDICT, and R. W. JACKSON. 1956. Antibiotics against plant disease. II. Effective agents produced by Streptomyces cinnamomeus forma azacoluta f. nov. Phytopathology 46: 575-581.
6. SMALE, B. C., and J. W. MITCHELL. 1960. Control of lima bean downy mildew with phenacridane chloride and closely related compounds. Plant Disease Reptr. 44: 684-686.
7. SMALE, B. C., O. L. SHOTWELL, T. G. PRIDHAM, and J. W. MITCHELL. 1958. Unpublished data.
8. TAKITA, T. 1959. Studies on purification and properties of phleomycin. J. of Antibiotics, Ser. A. 12: 285-289.
9. TAKITA, T., K. MAEDA, and H. UMEZAWA. 1959. Studies on phleomycin. J. of Antibiotics, Ser. A. 12: 111.

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STUNT OF SMALL GRAINS, A NEW DISEASE CAUSED BY THE NEMATODE TYLENCHORHYNCHUS BREVIDENS

Kenneth R. Langdon, F. Ben Struble, and H. C. Young, Jr.[1]

Summary

The nematode Tylenchorhynchus brevidens has been found associated with stunt symptoms of small grains in Oklahoma. Stunt symptoms were characterized by a general stunting and yellowing of aboveground plant parts, reduced tillering, and a reduction in grain yields. Root systems were typically stunted and darkened. A species of the fungus Olpidium also has been associated with stunt symptoms. In the greenhouse, stunt symptoms were reproduced on wheat in the presence of either T. brevidens or Olpidium sp. These two organisms in combination resulted in more severe symptoms than with either organism alone. Symptoms with T. brevidens were induced only after wheat had been vernalized; wheat grown with T. brevidens at constant temperatures was not stunted. This nematode has not previously been reported as a pathogen.

This nematode survived and reproduced on several different plant species including small grains, cotton, cucumber, and corn.

INTRODUCTION

Numerous yellowed, stunted areas were noted in wheat and barley nursery plots in the spring of 1958 on the West Farm of the Department of Agronomy, Oklahoma Agricultural Experiment Station, Stillwater. Preliminary examination revealed no evidence that the stunting was due to fungus, bacterium, virus, insect, or environmental factors. Soil samples from the stunted areas yielded high populations of the nematode Tylenchorhynchus brevidens Allen, 1955. This nematode was suspected of being the primary cause of the damage or at least of contributing to it. Other suspected plant parasitic nematodes, when present, were recovered only in trace quantities. Since T. brevidens had not previously been demonstrated to cause the observed damage, studies were initiated on the relationship of this nematode to the problem of stunting.

Beyond the original identification and description of this nematode little has been done. One study of its association with wheat (6), and a few observations of host range have been made (4, 5, 6, 7). Allen (1), in his description, noted grass as the type host for T. brevidens. Krusberg and Hirschmann (5) found T. brevidens in Peru associated with barley, potato, onion, alfalfa, garlic, carrot, and fruit trees but made no study of pathogenicity on these crops. Jenkins, et al. (4) in Maryland reported T. brevidens associated with alfalfa, barley, clover, grasses, oats, peas, timothy, and wheat. No great numbers of this nematode were found and no symptoms were associated with its presence.

Norton (6) found T. brevidens associated with wheat, oats, barley, and several grasses in Texas. He obtained evidence from greenhouse experiments that it was parasitic on wheat roots, but there was no evidence either from the greenhouse or the field that it was pathogenic. He concluded that T. brevidens was not a serious factor in the wheat root rot problem in Texas, but that with certain, undefined, conditions it could cause damage to wheat.

Vanterpool (7) has reported the presence of Asterocystis radicis de Wild. (Olpidium brassicae (Wor.) Dangeard) in the roots of cereals, particularly oats, in Canada. It caused root necrosis and yellowing of seedling leaves in his experimental plants. He cited reference to yellowing and scorching of the leaves of various cereals in Europe which had been attributed at least partially to infection by this fungus.

MATERIALS AND METHODS

Populations of T. brevidens were established on Concho wheat in the greenhouse with infested field soil. The nematode was maintained in the greenhouse by adding portions of infested soil with predetermined approximate numbers of T. brevidens to steamed soil and growing Concho wheat at about 70° F. This method yielded 80 to 90% T. brevidens in mixed nematode cultures. The remaining nematodes in these cultures were saprophytic forms; other than T.

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brevidens, known or suspected plant parasitic forms were never found in these samples. Populations of T. brevidens as high as 11,600 per half pint of soil were obtained by this method.

Preliminary tests confirmed Norton's (6) report that after a month there was a decline in T. brevidens populations on wheat in the greenhouse. A period of 2 months or more was necessary to obtain appreciable increases in nematode populations, consequently, all experiments were run at least 2 months.

Cultures eventually were established by hand picking several lots, each of 100 T. brevidens, and adding these to pots of steamed soil in which wheat was then planted. However, for all experiments reported requiring T. brevidens, portions of soil from stock cultures were mixed with steamed soil. For most tests infested soil was added, in order to obtain approximately 200 T. brevidens per pot. Preliminary experiments demonstrated this to be a near optimum inoculum level; with fewer numbers the nematode survived at a low level or required long periods for increase.

Nematodes were removed from soil samples by the sieving-Baermann technique (3) at the beginning of this study. For most of the work, however, a modification of Seinhorst's inverted-flask technique as described by Chapman (2) was used. This latter method gave more reliable and consistent results. All nematode counts, unless otherwise indicated, were made from half-pint soil samples.

RESULTS

Field Observations: Stunting and yellowing of wheat and barley were first associated with T. brevidens in the spring of 1958. In every case investigated this nematode was recovered from the soil in affected areas in such numbers as to make it suspect as a pathogen.

The small grains plots were routinely rotated to another area of the farm in 1959. In the area where Harbine barley was being increased, traces of stunting in spots were evident shortly after active growth resumed in the spring of 1959. Severe stunting was evident by March 7 and at this time soil samples were taken. Yields of T. brevidens were: from within a stunted area 400, from the periphery of this area 1060, and from an adjacent apparently normal area 480. Wheat plots in the same field also had stunted spots. From these, T. brevidens was recovered at a level of 690 and 350 for stunted and apparently healthy areas, respectively. By June 9 populations in this same wheat plot had increased to 1900 in the stunted and 700 in the apparently healthy area. Stunted spots in both crops increased little, if any, in size, but stunting increased in severity with time.

Aboveground symptoms associated with the presence of T. brevidens were a general stunting of the plants in spots ranging from about a foot up to several feet in diameter, an overall yellowish appearance, a pronounced yellowing and dying of lower leaves, fewer tillers, and fewer and smaller kernels. Symptoms below ground were shorter roots and fewer branch roots. The tips of many roots of affected plants were stubby in appearance and tissues were differentiated much nearer the tip than in healthy roots. Root symptoms are illustrated in Figures 1 and 2. Roots of affected plants were usually darker in color than those of healthy plants. Yield of grain from stunted wheat was 12.4 bushels/acre as compared with 34.0 bushels/acre from apparently healthy wheat. Symptoms generally were more severe in barley than in wheat.

Grossly the symptoms just described are similar to those associated with yellowing caused by Olpidium, and a species of this fungus was found in roots in nearly all stunted spots in wheats, oats, and barley. The level of infection in wheat was generally low and was considered of doubtful importance. The level of infection in barley was higher than in wheat and was generally correlated with the severity of stunting. The same was true in oats except more variation in varietal reaction was noted.

Soil and roots of various oat hybrids and varieties were checked for T. brevidens and Olpidium. Soil from a plot of Bronco yielded 200 T. brevidens per half pint; no Olpidium was found in the roots and no stunting was observed. A selection of the hybrid Forkedeer x Minnesota 0-363-3 showed moderate stunting. No T. brevidens were found, but the level of infection by Olpidium was quite high. Stunting in this case was attributed to effects of Olpidium. The most severe stunting was found in a plot of a selection of the hybrid Clintland x Mustang. The population of T. brevidens averaged a rather high 700 per half pint of soil; there was a moderate level of infection with Olpidium, about half that in the previous selection.

Since 1958, T. brevidens has been found in association with stunting in wheat and barley from several widely scattered locations in Oklahoma. Affected spots have ranged from a few feet in diameter up to an area involving about 5 acres. Wheat yield in this latter area in 1960 was 38.89 bushels/acre compared with 47.06 bushels for an adjacent apparently healthy area.

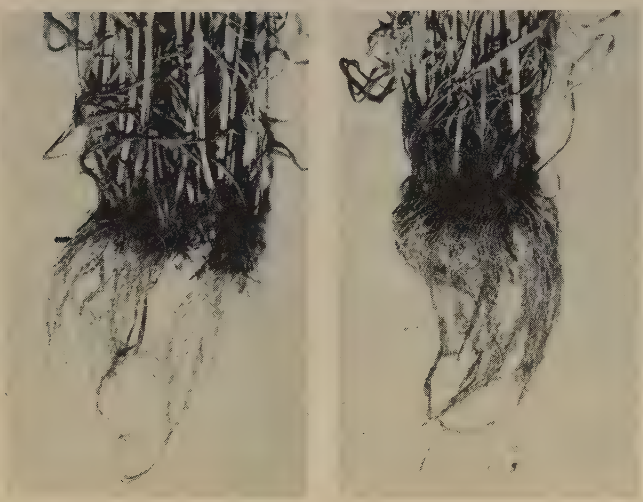

FIGURE 1. Comparison of crowns and roots of stunted (left) and healthy (right) mature wheat plants.

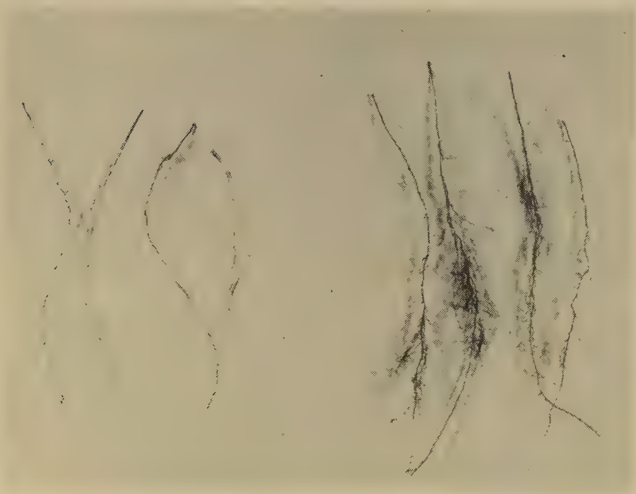

FIGURE 2. Comparison of individual roots of stunted (left) and healthy (right) wheat plants.

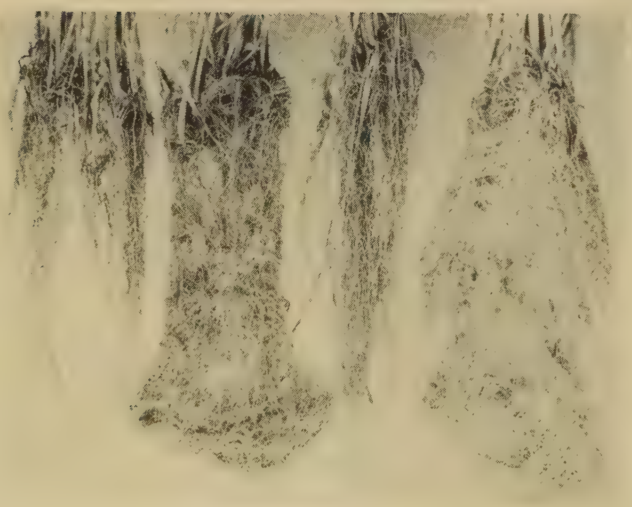

FIGURE 3. Comparison of root systems of wheat plants grown for 110 days with (left to right) T. brevidens plus Olpidium, Olpidium alone, T. brevidens alone, and control in steamed soil. Note differences in color of roots as well as in root volume.

Test weight was reduced from 58.0 pounds/bushel in the healthy area to 51.6 in the stunted area. There was some indication from the 1960 data that wheat yield and test weight were reduced even where stunting was not evident. Recovery of T. brevidens from soil in the healthy area was at the average rate of 374 as compared with 634 from the stunted area.

Pathogenicity Trials: A test was made to try to reproduce, in the greenhouse, symptoms observed in the field and to compare the relative pathogenic effects of T. brevidens and Olpidium sp. Four treatments with three replicates each were used as follows: 1) uninoculated control, 2) 400 T. brevidens per 6-inch pot, 3) 4 g of chopped Olpidium-infected roots per pot, and 4) a combination of nematodes plus Olpidium in the same amounts as in treatments 2 and 3. Concho wheat was planted and grown at 70° F for 2 weeks, vernalized at 40° for 45 days, and returned to 70° until the plants reached the boot stage (another 52 days). The most striking stunting of plants in the field has been observed at the boot stage.

Significant differences in height of plants in the various treatments were found. The inoculated plants averaged 35 cm in height compared with 32 cm for plants in soil with T. brevidens, 28 cm for plants in soil infested with Olpidium, and 26 cm for plants in soil with nematodes plus Olpidium. Striking differences also were noted in the root systems of these plants (Fig. 3). In the presence of T. brevidens stunting of the root systems was more evident than with Olpidium alone; top growth was reduced more with either Olpidium alone or in combination with the nematode. These results demonstrate that either T. brevidens or Olpidium alone can cause stunting and that the additive effect of both results in more severe stunting than with either one alone.

Wheat grown constantly at 70°F in soil to which T. brevidens had been added showed no effects of stunting.

Host Range Studies: The crops and varieties listed in Table 1 were evaluated as possible hosts for T. brevidens. Each variety was tested in three replications in 4-inch pots. Approximately 200 T. brevidens were added to each pot. Plants were grown at about 70° F for 2 months. The average number of T. brevidens recovered per pot with each crop variety and

Table 1. Suitability of selected hosts for reproduction of T. brevidens.

Crop	Variety	Average number T. brevidens per pot[a]	Rating[b]
Oats (Avena sativa)	Clintland x Mustang F_4 Sel. No. Stw. 583903	1494	S
Wheat (Triticum aestivum)	Wichita	1095	S
Oats	Forkedeer	771	S
Barley (Hordeum vulgare)	Harbine	743	S
Wheat	Concho	734	S
Rye (Secale cereale)	Balbo	657	S
Cotton (Gossypium hirsutum)	Stoneville 62	526	S
Cucumber (Cucumis sativus)	Marketer	505	S
Corn (Zea mays)	Golden Cross Bantam	465	S
Cowpea (Vigna sinensis)	Mississippi Crowder	284	I
Castor bean (Ricinus communis)	Hybrid 415	205	I
Sorghum (Sorghum vulgare)	Redlan	149	I
Tomato (Lycopersicon esculentum)	Sioux	131	I
Control[c]		95	U

[a] Recovery after 2 months from initial population of 200 per pot.
[b] S= satisfactory host on which there was a significant increase in nematode population; I= intermediate host with nematode population change not significant; U= unsatisfactory situation with a significant decrease in nematode population.
[c] Soil and nematodes with no plants.

the suitability of the variety as a host are shown in Table 1. Nematode increase was significantly different between the two oat varieties but not between the two wheat varieties.

This test suggests that the host range of this nematode is quite wide. Reproduction was generally greater on small grains than on most other crop plants. No plant was found in this study which supported fewer nematodes than soil with no plants at all.

Control Studies: A field test using two treatments and a control with four replications each was set up in the fall of 1958 in an area where T. brevidens had been a problem. One treatment was the nematocide ethylene dibromide (Dowfume W-85) at the rate of 6 gallons/acre overall; the other was composed of two fungicides, pentachloronitrobenzene (Terraclor) at the rate of 11 1/4 pounds/acre and a separate application of tetramethylthiuram disulfide (thiram) (Arasan) at the rate of 3 1/4 pounds/acre. The plots were planted to Concho wheat. The wheat was planted late and poor growth resulted during the winter and early spring. While no stunting was evident on any of the plots, there was an appreciable reduction in T. brevidens where ethylene dibromide was used. Soil samples were taken in June 1959 and 0, 330, and 680 T. brevidens were obtained for nematocide plots, fungicide plots, and control respectively. Unless late emergence and poor growth of the wheat were factors, failure to obtain stunting in at least some of these plots cannot be explained.

DISCUSSION AND CONCLUSIONS

While it has been shown that T. brevidens as well as a species of Olpidium can be responsible in causing certain stunting in small grains, no claim is made that only these organisms are responsible for stunting. Further studies are needed on the exact role of each of the organisms that may be involved in stunting.

Stunt symptoms induced by T. brevidens with wheat in the greenhouse were obtained only after the wheat had been vernalized. Whether these are the only conditions under which stunting can be induced has not been determined. The fact that stunt symptoms were not induced with wheat grown at a constant temperature suggests that environment is as important as the pathogen in disease development. Whether environment, particularly temperature, affects the host, the pathogen, or both, needs to be studied.

Stunting has been observed in the field only after winters that have been rather consistently cold and with adequate soil moisture. The influence of a more open type of winter on stunt symptom development and severity has yet to be determined. Norton (6) reported that field populations of T. brevidens were directly correlated with rainfall. Relatively dry conditions in Oklahoma for several years prior to the wheat-growing season of 1957-58 may provide at least a partial explanation as to why this nematode was not previously noticed as a problem on small grains.

Literature Cited

1. ALLEN, M. W. 1955. A review of the nematode genus Tylenchorhynchus. University of California Publ. Zool. 61: 129-166.
2. CHAPMAN, RICHARD A. 1958. An evaluation of methods for determining the number of nematodes in soil. Plant Disease Reptr. 42: 1351-1356.
3. CHRISTIE, J. R., and V. G. PERRY. 1951. Removing nematodes from soil. Proc. Helminthol. Soc. Wash. D.C. 18: 106-108.
4. JENKINS, W. R., D. P. TAYLOR, R. A. RHODE, and B. W. COURSEN. 1957. Nematodes associated with crop plants in Maryland. Maryland Agr. Exp. Sta. Bull. A-89.
5. KRUSBERG, L. R., and HEDWIG HIRSCHMANN. 1958. A survey of plant parasitic nematodes in Peru. Plant Disease Reptr. 42: 599-608.
6. NORTON, DON C. 1959. Relationship of nematodes to small grains and native grasses in north and central Texas. Plant Disease Reptr. 43: 227-235.
7. VANTERPOOL, T. C. 1930. Asterocystis radicis in the roots of cereals in Saskatchewan. Phytopathology 20: 677-680.

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REDUCTION OF FROST INJURY IN CRANBERRIES BY FUNGICIDE TREATMENTS[1]

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Abstract

Fungicide treatments resulted in a significant reduction in the amount of frost damage to cranberries. Since the freezing point of the berries was not affected by the treatments, it is possible that the effects of fungicides on the berries' environment may explain these findings.

Each fall a portion of the cranberry crop is frozen prior to harvest. Damage has varied from an estimated 60% in Massachusetts in 1917 (2) to a relatively small percentage of the crop in other years. However, the histories of certain bogs show that losses are commercially significant in many years. These bogs are susceptible to fall frost damage for many reasons, most important being the location of the bog in a "cold pocket" or lack of water to flood the bog to protect the berries from freezing (1). Freezing of cranberries results in disruption of the parenchymatous tissues and the consequent softening of the berry. Frozen berries usually become sticky and are difficult to sort from sound fruit (2).

In the fall of 1959 cranberries of the Howes variety treated with N-trichloromethyl thiophthalimide (Phaltan) were compared with untreated berries from an adjacent section. Only 0.1% of the berries in the fungicide-treated lot were frozen, whereas 9.3% of the untreated lot were frozen. Shortly after this observation was made, a grower reported that a similar condition existed on a number of his bogs which had been treated with zineb. These observations indicated that further study of the relation of fungicide treatment to reduction of frost injury in cranberries was desirable.

MATERIALS AND METHODS

An experiment had been initiated in 1955 in a 5 x 5 latin square design to evaluate the effects of four fungicides on Howes cranberries[2]. A similar series was established in an area in which cranberries of the Early Black variety were growing. Each of the four fungicides was applied twice to five randomized plots within the square. Five plots were left untreated. The first application was timed to coincide with the opening of approximately 5% of the cranberry flowers and the second was 10 to 14 days later. The chemicals were applied at the rate of 9 pounds/300 gallons water/acre/application, with the exception of Bordeaux mixture which was used at 30 pounds $CuSO_4$, 12 pounds agricultural lime/300 gallons water/acre/application. The fungicide treatments were repeated yearly in the same manner through the 1960 season.

In 1959 berries from one-third of each plot in the Howes variety experiment were exposed on a night in which the minimum temperature was 21°F. These berries were harvested the following day and the number of frosted ones tallied. In 1960 the test was repeated in the same manner, with the exception that the berries were exposed on two nights in which frost occurred. On the first night the minimum temperature was 23° and on the second it was 19°. In 1960 berries from both the Howes and Early Black experiments were included in the test. The average sample size from each plot was 321 berries, and samples ranged in size from 193 to 455 berries. The freezing points of treated and untreated berries were determined with a 12-point recording potentiometer. A thermocouple was inserted into each berry and the berries then placed in the freezing compartment of a refrigerator where they were held until they froze. In both 1959 and 1960 berries from each of the fungicide treatments and from the untreated plots were tested.

The work of Whitman (3) suggested that satisfactory freezing points of specimens under 2 1/4 inches in diameter could not be obtained without special preparation. For this reason a series of fungicide-treated and untreated berries were prepared for freezing in the same manner in which Whitman treated his cranberry samples. The cranberries were first cut into small pieces, placed in aluminum foil bags 2 1/4 inches in diameter and squeezed to eliminate

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[1]Contribution No. 1291 of the University of Massachusetts College of Agriculture Experiment Station, Amherst.

[2]Manganese ethylene bisdithiocarbamate = maneb, zinc ethylene bisdithiocarbamate = zineb, ferric dimethyl dithiocarbamate = ferbam. Bordeaux mixture (10 pounds $CuSO_4$, four pounds $Ca(OH)_2$, 100 gallons water).

air spaces. A thermocouple was then inserted to a depth of more than 1/2 inch into the sample and the freezing point determined.

RESULTS

The percentages of frozen fruit in the fungicide-treated and untreated plots are given in Table 1. Very little variation occurred between replicates within treatments. The one exception to this was in 1959 when berries from one ferbam plot contained 9.1% frosted berries. The

Table 1. Percentages of frozen cranberries in plots treated with fungicides as compared with frozen cranberries in untreated plots[a].

Variety	Year	Maneb	Zineb	Bordeaux	Ferbam	Untreated
Howes	1959	1.5**	0.7**	3.0*	3.7	7.6
Howes	1960	9.0**	12.9**	13.8*	14.2*	21.9
Early Black	1960	6.3**	14.6*	11.9*	12.5*	23.7

[a] Each figure represents the average percentage of frozen fruit in five plots.
* Significant at the 5% level over the untreated plots.
** Significant at the 1% level over the untreated plots.

plots treated with maneb, the most efficient cranberry fungicide currently used in Massachusetts, consistently contained fewer frosted fruit than plots treated with Bordeaux mixture or ferbam. These results show that fungicide treatments are related in some manner to the smaller incidence of frost damage in cranberries.

When Whitman's procedure for determining the freezing points of cranberries was followed, the cranberries froze at 29° to 30° F. These results were in accord with Whitman's findings (3). When the thermocouple was inserted directly into the whole berry, the freezing temperatures averaged 22°. Since it was not the purpose of the current experiments to evaluate methods for obtaining freezing points of cranberries, both methods were employed. Neither method revealed differences between freezing points of fungicide-treated and untreated berries.

DISCUSSION

Since fungicide treatment did not result in an alteration of the freezing point of the berry, it is proposed that the effects of fungicide treatment on the environment immediately around the berry may be important. The disease protection afforded by the chemical may result in a heavier leaf cover in the treated plots. The heavier leaf cover could, by entrapping radiant heat, give a degree of protection against frost damage. At present, experimental data to support this theory are lacking.

Literature Cited

1. FRANKLIN, H. J. 1948. Cranberry growing in Massachusetts. Massachusetts Agr. Exp. Sta. Bull. 447: 21-23.
2. FRANKLIN, H. J., H. F. BERGMAN, and N. E. STEVENS. 1943. Weather in relation to cranberry culture. Massachusetts Agr. Exp. Sta. Bull. 402: 36-37.
3. WHITMAN, T. M. 1957. Freezing points of fruits, vegetables and florist stocks. U.S. Dept. Agr. Marketing Research Rept. 196.

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PEAR DECLINE TRENDS IN WASHINGTON ORCHARDS[1]

L. P. Batjer, E. S. Degman, and N. R. Benson[2]

Abstract

The pattern of pear decline development on an individual tree basis was determined in 30 Washington orchards over a period of 4 years. Pear trees on oriental rootstocks had a much higher incidence of pear decline than those growing on either domestic seedlings or imported French. Most trees on oriental stock became progressively worse from 1956 to 1960. Trees in orchards on domestic seedling stocks had somewhat less decline than trees on imported French. In general, trees on either of these stocks did not become worse; on the contrary, most trees improved during the course of the survey. The possibility that many of the trees on domestic or imported French rootstocks were not suffering from bud-union decline is discussed. The effect of rootstocks on trees 2 and 3 years of age was similar to that obtained with older trees.

In 1955 when pear decline (3) became a serious threat to the pear industry of Washington, a number of questions arose concerning the future of existing orchards. Among these questions were the following: Will decline increase in orchards where only a few trees are affected with the disorder? If so, how rapidly will it spread? Will it eventually appear in orchards where it does not presently exist? Will trees showing decline symptoms recover? If not, how long will they linger? What effect does rootstock have on the behavior of pear decline? (It has been noted that the disease was more prevalent on trees with oriental rootstocks (1)). Can young trees be planted in the place from which a decline tree has been removed? The present study was designed to obtain information on some of these questions.

MATERIAL AND METHODS

In 1956, 30 typical Bartlett and D'Anjou orchards were selected in Yakima County, Washington for study of the occurrence of pear decline on individual trees. In each of these orchards a block of 60 to 100 trees was labeled and each tree was rated according to its condition in one of five classes. These classes are described as follows: 1, normal foliage with vigorous growth; 2, normal foliage but less shoot growth than 1; 3, very little terminal growth, with spur and shoot leaves smaller than on normal trees; 4, almost no terminal growth, small sparse foliage; 5, no terminal growth, very small and sparse spur leaves, and some trees nearly dead. Later a sixth class was used to denote trees that had either died or had been removed by the owner during the course of the survey because of their devitalized condition. All trees were rated 4 to 5 weeks after full bloom of each year from 1956 to 1960 inclusive. Tree age varied from 5 to 60 years: 11 orchards with trees on oriental rootstock (principally _Pyrus serotina_ or _Pyrus ussuriensis_) were about 30 years old, 10 on French seedlings (_Pyrus communis_) were approximately 60 years old, and 9 on domestic seedling rootstock (_Pyrus communis_) varied from 5 to 25 years old.

An experimental planting of 130 Bartlett pear trees on four different rootstocks was set out at the Tree Fruit Experiment Station, Wenatchee, Washington. About an equal number of trees of each rootstock were planted in the springs of 1958 and 1959 in single tree randomized plots. Growth performance on an individual tree basis was obtained in 1960.

RESULTS

The yearly growth status of each tree (Yakima County orchards) was compared with the condition of that tree in 1956. All trees on each rootstock were consolidated into one group in order to summarize the changes occurring. These summaries (Table 1) show the percentage change of all trees from the initial rating (1956) to that of 1958 and 1960. It is recognized that

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[1]Scientific Paper No. 2084, Washington Agricultural Experiment Stations, Pullman, Washington.
The authors acknowledge the aid of C. G. Woodbridge and E. C. Blodgett of the Washington Experiment Station in rating pear trees in the Yakima County orchards.

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Table 1. Pear tree ratings in 1956 as compared with ratings of the same trees in 1958 and 1960.

Rootstock group and class in 1956	Trees rated	% of trees in indicated class in 1958						% of trees in indicated class in 1960					
		1	2	3	4	5	6	1	2	3	4	5	6
Imported French:													
1	359	74	22	3	1	0	0	83	12	4	1	0	0
2	260	43	42	13	2	0	0	46	34	18	2	0	0
3	171	25	39	23	7	2	3	25	37	25	11	0	3
4	49	5	17	29	24	9	16	7	27	17	16	0	33
5	12	14	21	30	14	14	7	14	22	7	7	0	50
Domestic seedling:													
1	344	88	9	3	0	0	0	72	17	3	8	0	0
2	225	47	48	5	0	0	0	41	49	10	0	0	0
3	113	41	37	19	3	0	0	44	20	32	4	0	0
4	31	54	0	32	14	0	0	89	9	2	0	0	0
5	2	0	0	0	0	50	50	0	0	50	0	0	50
Oriental:													
1	129	61	19	15	3	1	1	40	27	16	5	3	9
2	180	8	29	39	17	3	4	3	22	40	17	5	13
3	330	1	6	37	34	9	13	0	1	23	39	14	23
4	185	0	5	24	38	23	10	0	1	7	29	20	43
5	51	0	0	7	29	47	17	0	0	0	24	41	35

Table 2. Effect of rootstock on 1960 performance of Bartlett pear trees planted in 1958 and 1959.

Rootstock	No. trees planted 1958-59	% trees with quick decline	% trees rated 3 and 4
P. communis (French seedling)	49	0	13
P. communis (domestic seedling)	36	0	7
P. calleryana	36	3	34
P. serotina	36	35	57

tree age is confounded with rootstock, but it will be shown later that even young trees are about as susceptible to the disorder as older trees; therefore, it is believed that tree age is not a factor in pear decline.

Pear trees on oriental rootstocks in the Yakima County orchards (Table 1) had a much higher incidence of pear decline than those growing on either domestic seedlings or imported French rootstocks. Most trees on oriental stock became progressively worse from 1956 to 1960. A substantial number of trees on this stock rated normal or nearly normal (class 1 and 2) in 1956 had declined into the 3, 4, 5, and 6 classes by 1960. Also the percentage of trees in class 6 (those removed principally because of quick decline or extremely low vigor) increased markedly from 1958 to 1960.

Orchards with trees on domestic seedling stocks had somewhat less decline than those on imported French (Table 1). In general, trees on either of these stocks did not become worse from 1956 to 1960. On the contrary, most trees (those rated 3, 4, and 5 in 1956) improved during the course of the survey.

Young trees (2 to 10 years old) in the Yakima orchards which were obvious replants in older orchards were not rated until 1960. In most cases these trees (all on domestic seedling rootstock) were normal and vigorous. In 1960, 95% of the replants in orchards where the older trees were on oriental stock were rated normal or nearly normal (class 1 and 2) whereas only 15% of the older trees were so classified. Many of these replants occupied a position in the orchard where trees had died from decline.

The reaction of 2- and 3-year-old trees on sensitive rootstocks (Table 2) was similar to that of older trees (Table 1). This would indicate that tree age is not a factor in pear decline. Figure 1 relates the percentage of the trees showing early fall red coloration with rating class.

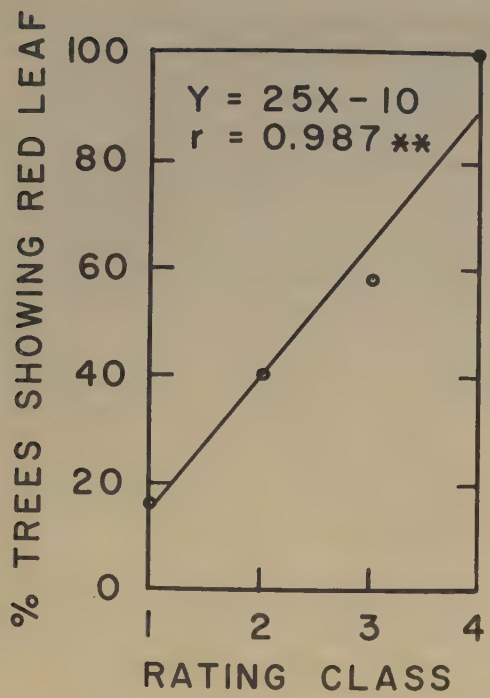

FIGURE 1. Relation between percentage of pear trees showing early fall red coloration and degree of pear decline.

All 130 trees including all four rootstocks (Table 2) were consolidated to make this figure. The equation is weighted according to the number of observations in each rating class. The figure shows that there is an increasing probability for the trees to have red foliage as decline becomes more severe. Blodgett and Aichele (2) found an association between decline and early red coloration on young trees.

DISCUSSION

In this study visible symptoms (small sparse foliage and lack of terminal growth) were used in determining the presence of slow decline. However, similar symptoms may be produced by factors such as girdling, "wet feet," malnutrition, winter injury, and drought. Unfortunately, pear decline has no macroscopic symptoms sufficiently specific for positive diagnosis. Batjer and Schneider (1) showed that this disorder usually is associated with a series of anatomical changes which are initiated by a necrosis of sieve tubes immediately below the bud union. With oriental stock they found a very close relation between apparent decline (as determined by macro-symptoms) and anatomical symptoms at the bud union. However, many of the trees growing on French seedlings were diagnosed negative (no sieve-tube necrosis at the bud union) even though they had visible symptoms characteristic of pear decline. Only one of 44 trees on domestic seedling stock sampled by Batjer and Schneider (1) showed sieve-tube necrosis.

In the present study a high percentage of the trees on imported French and domestic seedlings rated 3, 4, and 5 in 1956 had shown marked improvement by 1960. Thus, it would appear that many of these trees had recovered or were not afflicted with sieve-tube necrosis but that the low vigor was due to other causes. It should not be assumed that all the trees which were rated 3 or worse were suffering from bud-union decline (sieve-tube necrosis). As pointed out, low vigor may be caused by other factors and positive diagnosis of bud-union decline can be made only by anatomical examination. Likewise, "red leaf" in early fall is not a specific symptom of this disorder, but may also be associated with other causes that result in abnormal reduction in growth. However, "red leaf" is regarded as indicative when affected trees occur among otherwise vigorous, healthy trees in areas where pear decline is known to be present.

Literature Cited

1. BATJER, L. P., and HENRY SCHNEIDER. 1961. Relation of pear decline to rootstocks and sieve-tube necrosis. Proc. Am. Soc. Hort. Sci. 76: 85-97.
2. BLODGETT, EARLE C., and MURIT D. AICHELE. 1960. The association of a red-leaf condition of pear trees with pear decline. Plant Disease Reptr. 44: 904-907.
3. WOODBRIDGE, C. G., E. C. BLODGETT, and T. O. DIENER. 1957. Pear decline in the Pacific Northwest. Plant Disease Reptr. 41: 569-572.

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FURTHER OBSERVATIONS ON PEAR DECLINE IN WASHINGTON WITH PARTICULAR EMPHASIS ON QUICK DECLINE[1]

C. G. Woodbridge and E. C. Blodgett

In a previous paper the authors (5) reported some preliminary findings and the extent of "pear decline" in Washington. Now, 4 years later, a summary of subsequent observations is in order. The disease has spread down the Pacific Coast and is now in California (4). Certain trends with special emphasis on surveys of orchards made in the spring of each year are reported in a companion paper by Batjer, et al. (1). This report emphasizes the results of fall surveys of entire orchards or blocks of trees which have been observed annually since 1956. All of these orchards were in the Yakima Valley.

Anatomical studies of the phloem at the graft union have shown that slow and quick decline are different expressions of the same disease (2). As yet there is no answer to why some trees are affected by quick decline and others by slow decline. The resistance or susceptibility may be due to the use of seedling rootstocks. In the case of quick decline there are fewer normal roots but otherwise the degeneration of the root systems is similar. Also, in our previous paper (5) it was stated that oriental roots seemed to play an important role in decline. This observation has been confirmed. It has been shown that 1 to 3 year old trees on these rootstocks are highly susceptible to quick decline.

Recent work by Blodgett and Aichele (3) has shown that there is an additional syndrome, which has been termed "red leaf." This condition has been recognized for a number of years, but only recently has it been shown to be associated with decline (1, 3). The red-leaf condition is found on trees on oriental and communis type seedlings, both domestic (Bartlett) and imported French.

The number of trees affected by quick decline in seven orchards for the years 1956-1960 is shown in Table 1. In 1959 the orchards were not surveyed. With but a minor exception, the number of trees affected each year has decreased. In fact, no cases of quick decline were found in these orchards in 1960. Our 1956 report intimated that usually more vigorous trees were affected by quick decline. Results since then have shown that trees at any stage of slow decline may be affected. The slowing down in the rate of appearance of quick decline may be due to the fact that the more susceptible trees have died or have been removed. Weather conditions alone are not believed to account for this decrease. In 1960, our spring was late and cool but there was an unusually hot period in July. Hot weather does not appear to be a primary causal factor.

Table 1. The incidence of quick decline in some orchards in the Yakima Valley from 1956 to 1960.

Orchard no.	Total number of trees	Number of trees affected: 1956	1957	1958	1960
1	91	8	5	6	0
2	265	-	48	7	0
3	615	50	28	2	0
4	1169	138	113	41	0
5	1070	-	-	56	0
6	195	-	36	0	0
7	244	0	0	8	0

Age of tree does not seem to be an important factor in the occurrence of quick decline. In the orchards surveyed, the affected trees were 15 to 40 years old. However, in plots at Prosser and Wenatchee, 1- to 3-year-old trees on oriental (P. serotina, P. calleryana, and P. ussuriensis) root stocks were affected whereas trees on P. communis were not.

In one orchard 282 trees (24%) have been affected with quick decline since 1956. More than this number have been removed because of advanced stages of slow decline. Fortunately, the rate of spread is decreasing. Most of the pear replants, Bartlett on domestic seedling rootstocks, are doing well. Other replants, apples and peaches, are growing satisfactorily.

In another orchard, 18% of the trees in a block of 195 Bosc trees were affected with quick decline in 1957. Most of the trees appeared to have been growing vigorously. They were scat-

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tered throughout the block. Bartletts a few rows away in the same orchard were not affected. There has been no further incidence of quick decline. Slow decline has remained at a low level.

In 1958 a third orchard in which other experiments were being conducted suddenly took a turn for the worse. Besides typical decline, both quick and slow, 18% of the trees (190 of 1070) showed early defoliation and the few leaves that were left on October 8 were red rather than normal autumn yellow. The fallen leaves appeared to be of near normal size but the trees were rated, mainly on growth, as classes 3, 4, and 5 (5). In 1960 this orchard was mapped on September 20, two weeks earlier than in 1958. A large percentage of the leaves on the same trees were red and of normal size. However, again the growth of the trees was limited and they were assigned ratings as in 1958. This limited growth may be expected if the reddening is a symptom of decline. With early defoliation, the bark remains green and more healthy in appearance than in cases of more typical slow decline in which the leaves are small and pale throughout the growing season and the bark is greyish. This type of defoliation was mentioned in 1956 (5), but at that time it was thought to be more similar to quick decline than to slow decline.

Literature Cited

1. BATJER, L.P., E. S. DEGMAN, and N. R. BENSON. 1961. Pear decline trends in Washington orchards. Plant Disease Reptr. 45: 255-257.
2. BATJER, L. P., and HENRY SCHNEIDER. 1960. Relation of pear decline to rootstocks and sieve-tube necrosis. Proc. Am. Soc. Hort. Sci. 76: (In press).
3. BLODGETT, EARLE C., and MURIT D. AICHELE. 1960. The association of a red-leaf condition of pear trees with pear decline. Plant Disease Reptr. 44: 904-907.
4. NICHOLS, CARL W., HENRY SCHNEIDER, H. S. O'REILLY, THOMAS A. SHALLA, and W. N. GRIGGS. 1960. Pear decline in California. The Bulletin, Dept. of Agr., State of California. 49: 186-192.
5. WOODBRIDGE, C. G., E. C. BLODGETT, and T. O. DIENER. 1957. Pear decline in the Pacific Northwest. Plant Disease Reptr. 41: 569-572.

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THE TRANSLOCATION OF ROOT-APPLIED STREPTOMYCIN IN BEAN[1]

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Abstract

The bark of Pinto bean stems accumulated more streptomycin than did the wood when the antibiotic was introduced via the root solution. The wood: bark ratio of the quantity of streptomycin after 24-hour feeding periods ranged from 1:2 to 1:6. When each of the vascular tissues was selectively blocked, streptomycin was translocated upward predominantly in the xylem. The presence of the antibiotic in the bark indicates an exchange between the xylem and phloem.

Streptomycin has been effectively used in the control of certain plant diseases, and a number of studies have been done on the uptake and translocation of this antibiotic by various plant species (4, 5, 6, 7, 8).

However, there has been relatively little attention paid to the specific tissue systems involved in the transport of this and other antibiotics. It has been tacitly assumed that export from the leaf takes place in the phloem, and that upward transport through the stem, resulting from root absorption, is via the xylem transpiration stream. While these assumptions are logical in light of our knowledge of translocation patterns in plants, recent radioisotopic work has indicated that there is a significant exchange of materials between the xylem and phloem and bi-directional movement of materials in the phloem (1, 2).

Therefore, we decided to examine in more detail the translocation of streptomycin in bean.

MATERIALS AND METHODS

Bean plants, Phaseolus vulgaris "Pinto," were raised at 27°C in quartz sand, and watered with Hoagland's solution, which was modified by using 150 mg/l of Versenol F (Dow Chemical Company) as an iron source. When the first trifoliate leaves were just unfolding, uniform plants were selected, carefully uprooted, and washed. They were placed in beakers in which the roots were immersed in 200 ml of Hoagland's solution plus streptomycin sulfate at a concentration of 150 μg/ml. The root solutions were continuously aerated because preliminary experiments indicated that more streptomycin was present in the stem when the nutrient solution was aerated. During this streptomycin-feeding period the plants were kept in a controlled-environment chamber in which a 12-hour light period (24°C) alternated with a 12-hour dark period (18°C). Light was provided by combination of incandescent and fluorescent lights at 1200 f.c.

After various time periods, the plants were harvested and divided into parts. These were weighed, ground in a tissue grinder, and centrifuged. One-tenth ml aliquots of the supernatant solution were bioassayed for streptomycin content according to the method of Maier (5), employing a streptomycin-sensitive strain of Bacillus subtilis Cohn. The results were expressed as μg streptomycin sulfate per g of fresh plant tissue.

Two types of experiments were undertaken to determine what tissues are involved in streptomycin transport. The first involved separating the stem between the cotyledonary and the primary leaf nodes into bark and wood after the feeding period and bioassaying each portion separately. The second consisted of selectively "blocking" each of the vascular tissues prior to the feeding period; the phloem by steam-girdling the hypocotyl just below the cotyledonary node, and the xylem by making a vertical slit in the bark above the cotyledonary node, folding it back, and excising out a 1-cm portion of the wood underneath.. The slit was then covered with Saran Wrap and the plants immediately placed in a humid atmosphere. After the feeding period, the stem above the block was divided into bark and wood and each portion bioassayed separately. The term "bark" refers to all tissues external to the vascular cambium, while "wood" refers to all tissues internal to the cambium.

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RESULTS AND DISCUSSION

The results of a representative experiment in which the plants were fed for two time periods are presented in Table 1. As would be expected, the longer the feeding period the greater the accumulation of antibiotic in the stem. Of more importance is the fact that a significant portion of this accumulation was found to be in the bark and, by implication, in the phloem.

Table 1. Concentration of streptomycin in different plant parts after two time periods.

Plant part	Streptomycin concentration ($\mu g/g$ fresh tissue): 12-hour feeding period	24-hour feeding period
Wood (xylem)	63	64
Bark (phloem)	148	460
Petiole of primary leaf	17	144
Primary leaf	0	0
Stem above primary-leaf node	42	215
Trifoliate leaves	0	trace

Table 2. Effect of four light regimes on the streptomycin concentration of bark and wood.

Feeding period (in hours): light	dark	Streptomycin concentration ($\mu g/g$ fresh tissue): wood (xylem)	bark (phloem)
6	0	403	525
0	6	284	249
12	12	387	939
0	24	249	1471

Table 3. Effect of selective blocking of different vascular tissues on the translocation of streptomycin.

Treatment	Streptomycin concentration ($\mu g/g$ fresh tissue): wood (xylem)	bark (phloem)
Wood removed	trace	trace
Bark steam-girdled	1128	103

Among the various experiments, the ratio of the quantity of streptomycin in the wood to the bark after a 24-hour feeding period ranged from 1:2 to 1:6.

Similar results are shown in Table 2. The plants were subjected to various light and dark periods to determine how light affects translocation. For the 6-hour period, the effect of light was to increase the amount of streptomycin in the stem. However, no such effect was observable after 24 hours.

The results obtained when each of the vascular tissues was selectively blocked are presented in Table 3. The plants were analyzed for streptomycin after an 18-hour feeding period. When the phloem was blocked, streptomycin was still found in the stem above the block, whereas only a trace was found above the block when the xylem was intercepted. These results indicate that the presence of streptomycin in the bark is due primarily to a lateral movement from the xylem and, to a much smaller degree, to an upward movement in the phloem. It should be emphasized, however, that these conclusions may not be valid for an intact plant under natural environmental conditions.

These results agree with those of Dye (4) and Pramer (7, 8), who found that under somewhat similar conditions streptomycin was initially accumulated in the stem, with concentration decreasing from roots to apex. They suggested that such results could be explained by the hypothesis of Charles (3), that the positively charged streptomycin molecules are bound to the negatively charged surfaces of the xylem. We suggest that at least two additional processes have contributed to this distribution pattern, namely that: a) the rate of upward streptomycin translocation, in terms of the amount transported per unit time, is much higher in the xylem than in the phloem, and b) there is an exchange of the antibiotic between the xylem and phloem resulting in a higher concentration in the phloem.

Literature Cited

1. BIDDULPH, O. 1959. Translocation of inorganic solutes. Vol. II,

pp. 553-603. In Plant Physiology, ed. F. C. Steward. Academic Press, New York.

2. BIDDULPH, O., and R. CORY. 1960. Demonstration of two translocation mechanisms in studies of bidirectional movement. Plant Physiol. 35: 689-695.
3. CHARLES. A. 1953. Uptake of dyes into cut leaves. Nature 171: 435-436.
4. DYE, M. H. 1956. Studies on the uptake and translocation of streptomycin by peach seedlings. Ann. Appl. Biol. 44: 567-575.
5. MAIER, C. R. 1960. Streptomycin absorption, translocation, and retention in hops. Phytopathology 50: 351-356.
6. MITCHELL, J. W., W. J. ZAUMEYER, and W. H. PRESTON, Jr. 1954. Absorption and translocation of streptomycin by bean plants and its effect on the halo and common blight organisms. Phytopathology 44: 25-30.
7. PRAMER, D. 1953. Observations on the uptake and translocation of five actinomycete antibiotics by cucumber seedlings. Ann. Appl. Biol. 40: 617-622.
8. PRAMER, D. 1954. The movement of chloroamphenicol and streptomycin in broad bean and tomato plants. Ann. Bot. 18: 463-470.

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SUGGESTED CHANGES IN THE TERMINOLOGY OF PLANT DISEASE CONTROL

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The various methods employed in the control of plant diseases have been classified by Whetzel (1, 2) under the four principles: exclusion, eradication, protection, and immunization. Whetzel (2) included selection, hybridization, nutrition, medication, and vaccination under immunization. I contend that the terms denoting the first three principles convey the sense for which they stand but that the last term does not.

In my suggested scheme, plant disease control measures are subsumed under exclusion, eradication, protection, selection, and physiocommutation, the last two terms being used as a substitute for Whetzel's immunization.

This change in terminology is suggested because of the misleading connotation of the term immunization, which is borrowed from animal pathology. Immunization is a process of rendering an organism immune. Almost all plant pathologists consider immunity in plants to be absolute in nature, and that the term implies a condition in which a plant does not react physiologically to the irritations afforded by a microorganism or its products. For example, an elm tree is inherently immune to the wheat stem rust organism and does not exhibit a physiological or a morphological change when in association with this organism. The term resistance is used for any condition intermediate between susceptibility and immunity. It may be generalized, therefore, that immune plants exist naturally. Since I have found no reports of an otherwise susceptible plant being immunized by artificial means, it appears that the term immunization cannot correctly be used in plant pathology.

The naturally immune plants may, however, be selected for breeding purposes and the genes conferring natural immunity may later be transferred from the parents to the progeny. Since the natural immunity conferred by the transferred genes is an inherent quality, and since the process of selection involves the selection or development of a new phenotype, it surely cannot be included under immunization, as Whetzel suggests. Selection, therefore, is considered as a separate principle in the present scheme.

Because of the inapplicability of the term immunization to the physiological changes in plants rendered resistant to a disease organism by altered nutrition, by medication, and so forth, a new term physiocommutation has been coined. This term is derived from physis (Gr. = nature) and commutatus (L = alteration, change). Physiology in its broadest sense means the knowledge of nature, but as used in biology is concerned with the mechanics of function. Physio- has been used as a prefix denoting either physiological, as in physiopathology, or as physical, as in physiotherapy. Here the prefix physio- has been used to mean physiological. The term physiocommutation, therefore, means physiological alteration.

In the present scheme, the term vaccination used by Whetzel is to be replaced by protective inoculation because of the misleading connotation of the former. Vaccination, another term borrowed from animal pathology, implies the formation of antibodies, which have not been demonstrated in plants. Consequently, the term vaccination appears inapplicable, in its usual sense, to plant pathology.

In this suggested terminology an attempt has been made to avoid misconceptions that could arise from the prior ill-chosen terminology. For this reason the terms selection, physiocommutation, and protective inoculation are offered, the first two as substitutes for Whetzel's immunization and the last as a substitute for his vaccination. It is believed that the widespread use of this scheme would minimize erroneous conceptions resulting from the connotations inherent in some of the terms borrowed from animal pathology.

Literature Cited

1. WHETZEL, H. H. 1929. The terminology of phytopathology. Proc. Int. Cong. of Plant Science 2: 1204-1215.
2. WHETZEL, H. H. 1950. Principles of plant disease control. Class notes (1950 revision). Department of Plant Pathology, Cornell University, Ithaca, New York. 32 pp.

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LIMITATIONS OF THE HOT WATER IMMERSION TREATMENT FOR THE CONTROL OF PHYTOPHTHORA BROWN ROT OF LEMONS

L. J. Klotz and T. A. DeWolfe

Summary

For adequate control of brown rot of lemons in the packing house, an immersion of 4 or more minutes in the treating tank solution at 115° to 120° F is necessary. The treatment will arrest decay in lemons that had been infected as long as 60 hours previously if the temperature of the fruit had not gone above 54° before treatment. To avoid rind oil injury, cold turgid lemons should be wilted slightly before immersion in hot solution. The nearer the orchard temperature is to the optimum growth temperature of the fungus (78° to 80°) during and just following the winter and spring rains, the greater the number of infected lemons and destructiveness of brown rot. The rate of penetration and decay of lemons by the fungus is more rapid in mature than in immature fruit. Although cold solutions of fungicides -- such as a 1-1000 solution of copper sulfate or hypochlorite solutions -- were very effective against pure cultures of the brown rot fungus, they were ineffective in stopping decay.

An immersion of 4 or more minutes in water or in washing-treating solution at a temperature of 115° to 120° F is essential for the control of brown rot of lemons. However, cold, turgid lemons may release rind oil and show the surface injury called rind oil spotting (oleocellosis) if they are immersed in the hot solution of the treating tank soon after picking. To avoid this injury, such fruit must be kept for a period and allowed to wilt slightly before the immersion treatment. The length of the conditioning period required depends upon relative humidity or desiccating power of the air prior to washing and treating, and the degree of turgidity and tenderness of the fruit when delivered to the packing houses.

The longer the conditioning period required, the greater the danger of the development of brown rot in the lemons infected by zoospores in the orchard. The purpose of this report is to show how long after infection of the fruit by the swimming zoospores of the brown rot fungus the hot water immersion treatment is effective in stopping the development of the decay, and to point out the factors upon which the effectiveness of the treatment depends. The incubation temperature following infection, the stage of maturity of the fruit, and the rate of penetration by the fungus are the factors considered.

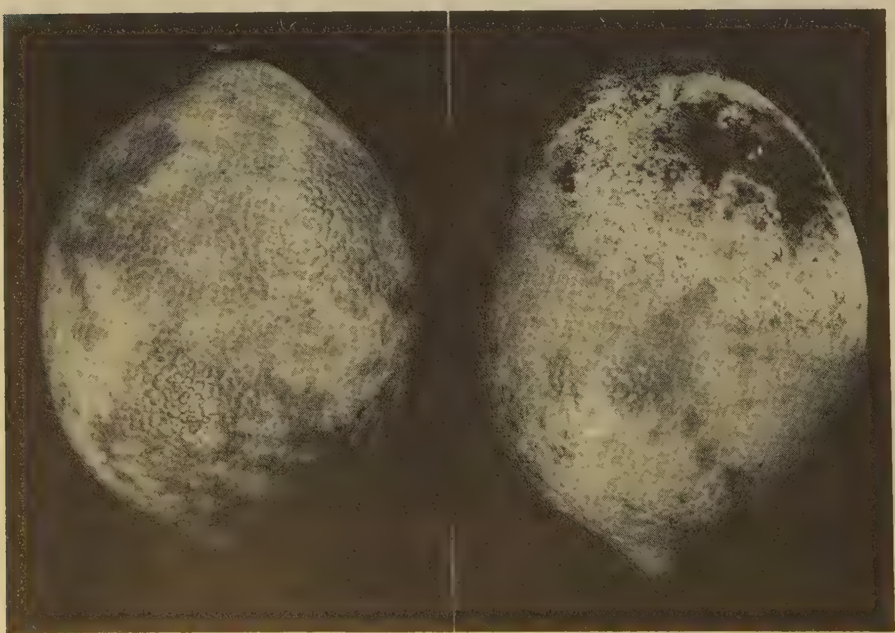


FIGURE 1. Left -- rind oil spotting (oleocellosis). Right-- rind oil spotting followed by green mold.

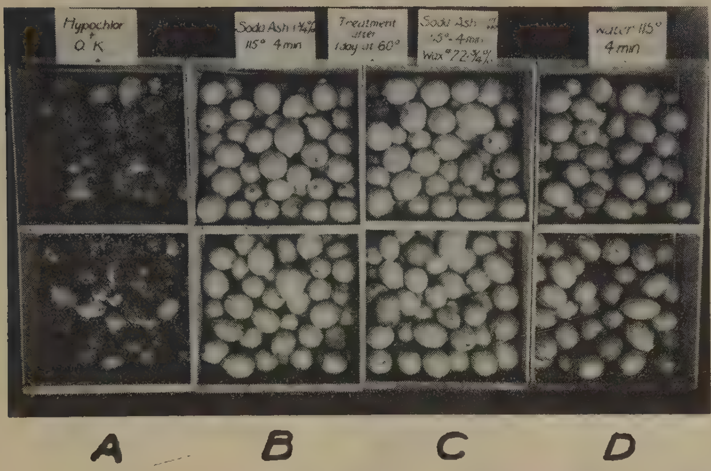


FIGURE 2. Brown rot control treatments: A, cold treatment (hypochlorite and sodium-o-phenylphenate in solution), all lemons decayed; B, C, D, hot treatments, only 2 lemons in 3 boxes decayed. All treatments 1 day after infection; fruit "held" (incubated) at 60° F during that period.

Table 1. Effect of incubation temperature and period on control of brown rot by immersion in 1-1000 copper sulfate ($CuSO_4.5H_2O$) at 77° F.

Incubation temperature		Hours incubated before treatment						
°C	°F	3	6	9	12	24	48	72
3	37.4	-	-	-	-	-	-	-
12	53.6	+	+	+	+	+	+	+
18	64.4	+	+	+	+	+	+	+
24	75.2	+	+	+	+	+	+	+
30	86.	+	+	+	+	+	+	+

+ = brown rot; 1 = no brown rot.

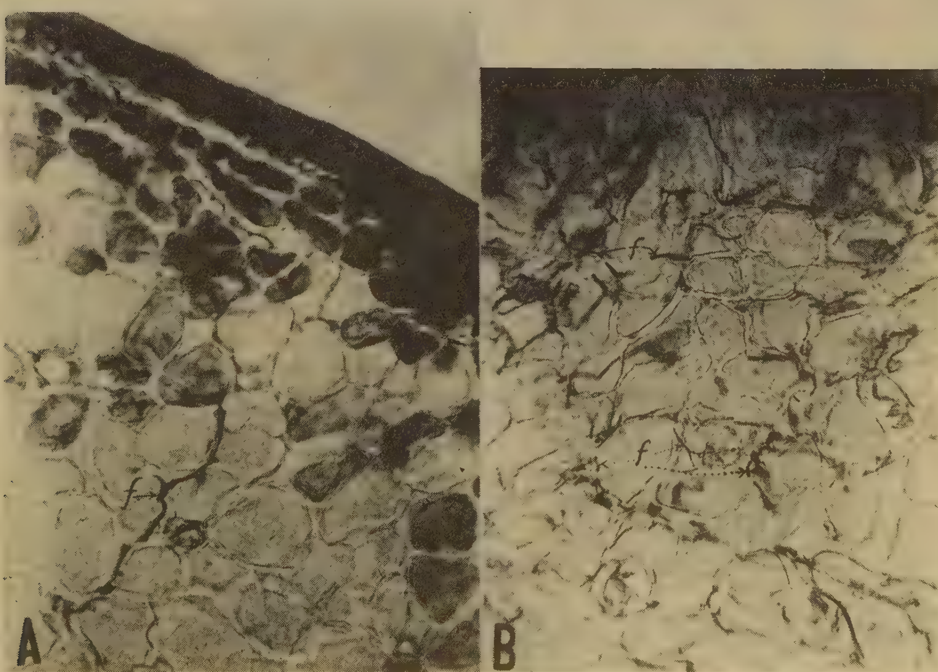


FIGURE 3. Penetration of brown rot fungus (f) into lemon rind: A -- fungus (f) at a depth of 818μ (.032 inches) after 72 hours at 54° F (about X 300). B -- complete involvement of rind (4873μ or .192 inches) after 48 hours at 72° F (about X 138).

Table 2. Effect of incubation temperature and period on control of brown rot by immersion in hot water (119° F, 4 minutes).

Incubation temperature °C	°F	Hours incubated before treatment: 3	6	9	12	24	30	36	42	48	54	60	72
3	37.4	-	-	-	-	-	-	-	-	-	-	-	-
12	53.6	-	-	-	-	-	-	-	-	-	-	-	+
18	64.4	-	-	-	-	-	-	-	-	+	+	+	+
24	75.2	-	-	-	-	-	+	+	+	+	+	+	+
30	86.	-	-	-	-	-	+	-*	+	+	+	+	+

+ = brown rot; - = no brown rot; * = green lemons.

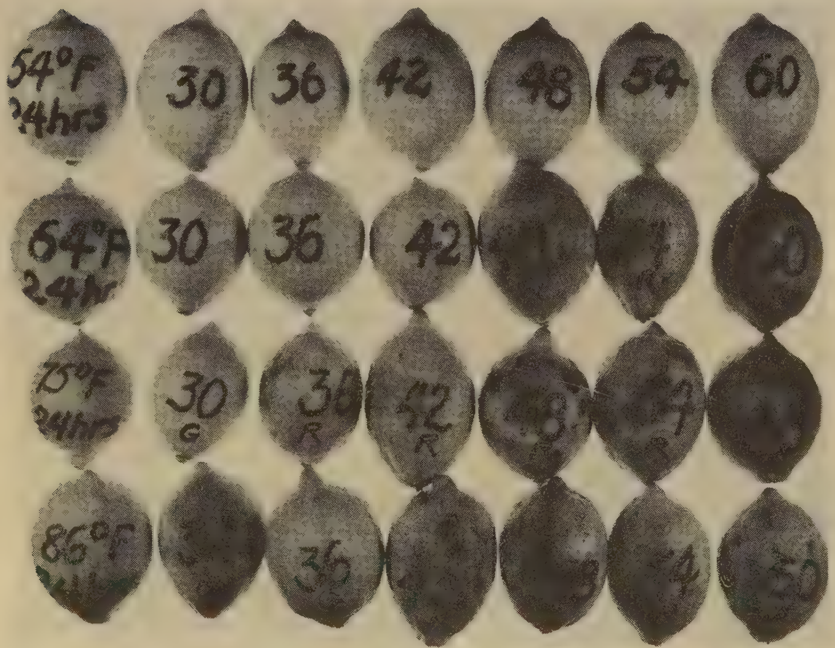


FIGURE 4. Effectiveness of hot water immersion treatment (118°-120° F for 4 minutes) in controlling brown rot of lemons. R means decaying; figures give number of hours' incubation before treatment or time elapsing between inoculation and treatment. Lemons of each horizontal row were incubated at temperature shown on left hand lemon of each row. G means green lemon.

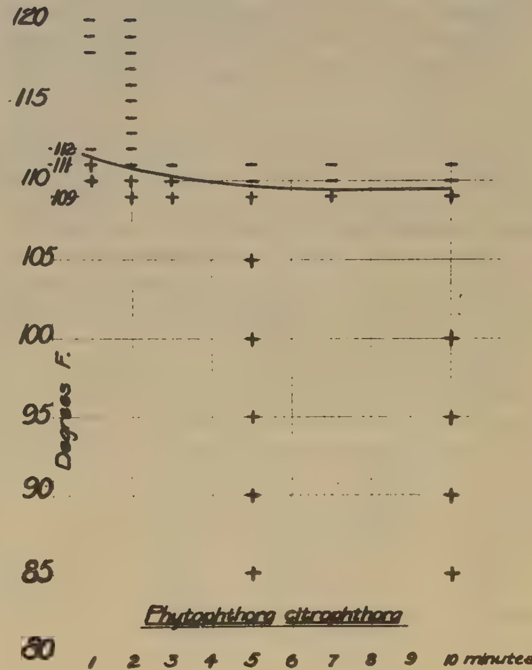
Table 1 and Figure 2 show the relative ineffectiveness of a cold toxic solution in preventing brown rot. In this experiment the lemons were placed for 24 hours in incubators at the temperatures shown, then inoculated by atomizing them with a zoospore suspension of the brown rot fungus and respectively incubating seven lots of fruits from each of the five incubators for the periods shown before immersing them in a 1-1000 solution of copper sulfate. While it has been shown that a 1-75,000 solution of copper sulfate is sufficient to kill the fungus, even the concentrated (1-1000) solution is ineffective after 3 or more hours at temperatures of 53.6° F and above. A temperature of 37.4° without the subsequent treatment was unfavorable for the development of brown rot, for only 5 of the 14 heavily inoculated lemons of the check lot at that temperature eventually developed the decay.

The reason for the failure of the cold copper sulfate or of hypochlorite solution to stop the decay is apparent when we consider that the fungus has penetrated the rind surface within 3 hours and then is beyond the reach of the fungicide. Figure 3, A and B, gives some idea of the rate of penetration. of the fungus into the rind. Once the organism is inside, only heat will reach and kill it. The lethal temperature exposures are shown in Figure 6.

Table 2 and Figures 4 and 5 show that the 4-minute hot water immersion treatment is effective in stopping decay even 60 hours after infection if the temperature of the lemons is 53.6° F or below. Since during rainy periods in California, when infection takes place, temperatures do not frequently exceed 53.6°, picked lemons could safely be kept up to 60 hours before treatment.

FIGURE 5. (left) Effect of the "holding" temperature or incubation temperature on hot water immersion treatment for control of brown rot.

FIGURE 6. (right) Temperature-exposures lethal to brown rot fungus: - means fungus killed; + means fungus not killed. Fungus (zoospores and mycelium) in water suspension in capillary tubes exposed to temperatures of water bath.



Immersions of less than 4 minutes are less effective because, with shorter exposures, the temperatures that kill the fungus do not penetrate as deeply into the rind and, as a result, the invading organism more frequently escapes. An exposure of 111° F for 2 minutes killed the fungus (Fig. 6). To demonstrate the rate of penetration of heat, experiments were conducted in a water bath at 118° using thermocouples inserted in the rind of immersed lemons (light and silver green) whose initial temperature varied from 36° to 95°. The tests with the initial temperature of the lemons at 60° revealed that in 1 minute a temperature of 111° had penetrated to a depth of 145μ (.0057 of an inch), in 2 minutes to 169μ (.006653 of an inch), in 3 minutes to 186μ (.00732 of an inch), and in 4 minutes to 207μ (.0082 of an inch). The higher the initial temperature of the lemon when it is immersed, the deeper the lethal temperatures penetrate the rind; for example, when the initial temperature was 78.2°, a temperature of 111° was attained at a depth of 318μ (.0125 of an inch) after 4 minutes' immersion in water at 118°. However, the nearer the temperature of the lemons is to the optimum temperature for growth of the fungus (78° to 80°), the deeper the parasite will penetrate the rind in a given time.

Only silver and a few light green and tree ripe lemons were available for these experiments. It has been repeatedly observed that the more immature the lemons the less rapid the penetration of the fungus and the development of brown rot. Accordingly, it is highly probable that if we had used green, immature lemons, we should have found the immersion treatment effective even after a longer period of incubation than 60 hours. Table 2 and Figure 4 also give an explanation for the great destructiveness of brown rot during relatively warm spring rains when the incubation temperatures are more favorable for rapid penetration of the fungus than during the winter months.

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BROWN ROT CONTACT INFECTION OF CITRUS FRUITS PRIOR TO HOT WATER TREATMENT

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Summary

Brown rot infections of sound citrus fruits which develop in picking boxes from contact with fruits infected in the orchard by zoospores may cause important losses. Such losses can be minimized by treatment in hot cleaning-fungicidal solutions as soon as the fruits have lost enough moisture to avoid liberation of their rind oil. The fruit rind surface must be slightly dried to tolerate the immersion in solutions at 46° to 49° C (115° to 120° F). Holding citrus fruits on the washroom floor longer than necessary will result in greater losses from brown rot.

Growers and processors know that Phytophthora brown rot is prevalent and causes important losses of citrus fruits during the rainy season (1, 2, 3, 5). The purpose of this report is to point out the importance of contact infections that develop during the period the fruits are in field boxes, that is from the time they are picked until they are dumped into the hot treating solutions in the packinghouse. Fruits with incipient, undetectable primary infections occurring in the orchard are frequently placed in picking boxes along with sound fruits (1). These infections develop into areas of decay at a rate dependent upon the temperatures to which they are exposed. Sound fruits in contact with the decaying fruits in turn become infected.

Results of the experiments reported here show the effects of time and temperature on the development of secondary or contact decay in sound citrus fruits as they stand on the packinghouse floor awaiting the washing and hot water treatment.

Eighty-four navel oranges with brown rot (3) were distributed among sound navels in four field boxes, thus, 21 infected fruits in each box. Two of the boxes were placed in a 10° C (50° F) room and 2 in a 20° C (68° F) room for 5 days. The original 84 infected fruits were then removed and observations for possible contact decay made on the following second, third, and fifth days. The number of contact decays that developed among the sound fruit are shown in Table 1. All fruits were held at room temperatures of 21° to 24° C (70° to 76° F) after removal of the decays. These oranges were picked on April 5, 1960, and were slightly overmature.

Table 1. Development of contact decay in navel oranges after contacting brown rotted fruits for 5 days at 10° or 20° C (50° or 68° F).

Box number	5-day incubation at:	No. of contact decays after removal of 21 decayed fruits from each box after: 2 days	3 days	5 days	Total contacts decayed	Number sound oranges remaining	% oranges decayed
1	10° C(50° F)	0	4	16	20	134	12.99
2	10° C(50° F)	0	5	14	19	132	12.58
3	20° C(68° F)	13	6	6	25	124	16.77
4	20° C(68° F)	14	5	4	23	125	15.54

To learn the effect of a wider range of incubation temperatures on development of brown rot from contact with infected fruits, six infected fruits were placed among sound fruit in each of seven large battery jars. These were covered with cheesecloth to exclude fruit flies, two-thirds covered with newspaper, and placed at the incubation temperatures shown in Table 2.

The two tests described indicate that where navel oranges are held on the floor for 5 days before treatment infection of sound fruits by contacting fruits with brown rot can cause important losses. Early stage infections may escape the heat of the treating tank and detection by the sorters and packers and thus the infected fruits go into the cartons and cause further contact decay during shipment and storage.

To learn the effect on incidence of contact decay of incubation periods of 1, 2, 3, 4, 5, 6, and 7 days at 10° C and 20° C (50° F and 68° F) the following procedure was followed. Eight field boxes of sound navel oranges (picked 4 and 5 January, 1961) and a box of 140 oranges decayed by the common brown rot fungus were placed in the 10° C (50° F) room and the same number in

Table 2. Development of contact decay in navel oranges after contact with brown rotted fruits for 5 days at seven incubation temperatures.

Battery jar number:	5-day incubation at:	No. of contact decays after removal of 6 decayed fruits from each jar after: 2 days	3 days	5 days	Total contacts decayed	Number sound oranges remaining	% oranges decayed
1	30° C(86° F)	1	1	0	2	25	7.41
2	27° C(80.6° F)	4	1	0	5	19	20.83
3	24° C(75.2° F)	2	0	1	3	21	12.50
4	21° C(69.8° F)	5	1	4	10	20	33.33
5	18° C(64.4° F)	9	1	1	11	16	40.74
6	15° C(59° F)	0	1	1	2	25	7.41
7	12° C(53.6° F)	0	4	0	4	23	14.81

Table 3. Effect of period of contact with brown rot fruits and temperature on development of contact decay in Washington Navel oranges.

Period of contact (incubation) days	Incubation temperature: 0° C	° F	Day contact decays found: 3rd	5th	7th	14th	Total decays after 14 days	Total sound fruits	% decays	Total decays (sum of all four incubation periods) Number	%
1	10	(50)	0	0	0	0	0	89	0		
2	10	(50)	0	0	0	0	0	105	0		
3	10	(50)	0	0	1	0	1	93	1.06		
4	10	(50)	0	0	0	1	1	93	1.06		
5	10	(50)	0	0	1	0	1	94	1.05		
6	10	(50)	0	0	1	0	1	98	1.01		
7	10	(50)	0	1	0	0	1	105	0.95	5	0.73
1	20	(68)	0	1	2	0	3	73	3.94		
2	20	(68)	0	1	2	2	5	91	5.21		
3	20	(68)	0	2	1	1	4	97	3.96		
4	20	(68)	5	3	3	1	12	85	12.37		
5	20	(68)	6	1	1	0	8	88	8.33		
6	20	(68)	3	3	4	0	10	87	10.31		
7	20	(68)	5	2	0	4	11	85	11.46	53	8.04

the 20° C(68° F) room to allow them to attain the temperature of the rooms. The next day 20 well marked rots were distributed among the sound oranges of each box. Every day for 7 days a box from each of the two temperature rooms was brought to the laboratory (21° -26° C or 70° -78° F) and the 20 primary rots removed (4). After 3, 5, 7, and 14 days in the laboratory only the originally sound oranges that had been in contact with the 20 primary rots and were marked with waterproof ink were examined for evidence of contact decay. To maintain the conditions existing in full boxes, the fruits not contacted were replaced in the boxes with those that had been contacted. The results are recorded in Table 3.

The advantage of keeping the fruits at the lower temperature before washing is seen from the small number of decays that developed -- only 0.73% following incubation at 10° C (50° F). At the higher incubation temperature of 20° C (68° F) losses ranged from 4 to 12%, with an average of 8.04%. The effect of hot solution immersion treatments in checking the contact infections that take place in picking boxes is shown by the following experiments with lemons.

Growers of lemons suffer the greatest losses from brown rot. The importance of temperature and of the period of time elapsing between picking and the hot solution treatment and the comparative effectiveness of two kinds of treatments were determined by the following experiments.

The incubation temperature procedure described for the second series of tests with navel oranges was employed with lemons. The primary brown rot lemons were removed from contact with the sound fruits after 1, 2, 3, 4, and 5 days' incubation at 10° C and 20° C (50° F and 68° F), respectively. Brown rot contacted lemons from two boxes from each of the two temperature rooms were randomized separately, 100 being used from each temperature for each of the two hot solution treatments. The lemons exceeding 100 were not treated but were kept for

Table 4. Influence of temperature and period of contact with decayed fruits on effectiveness of hot solution treatments for control of contact brown rot of lemons.

Days[a] in contact with brown rotted lemons	5 minutes 49°-47°C (120°-116°F) soda ash 2.5% Mermaid soap 0.5% Number or percent decayed	sound	3 minutes 49°-47°C (120°-117°F) sodium orthophenylphenate 0.375% (pH 12) Number or percent decayed	sound	Untreated decayed	sound	% decayed
Temperature during contact 10°C (50°F)							
1	0	100	0	100	2	51	3.77
2	0	100	0	100	0	33	0.00
3	0	100	0	100	0	55	0.00
4	0	100	3	97	7	54	11.47
5	1	99	2	98	13	37	26.00
8					33	207	13.75
Total	1	499	5	495	55	437	11.18
Temperature during contact 20°C (68°F)							
1	0	100	0	100	8	57	12.31
2	8	92	10	90	12	38	24.00
3	9	91	12	88	7	21	25.00
4	20	80	26	74	19	36	34.54
5	26	74	28	72	8	29	21.62
8					44	67	39.64
Total	63	437	76	424	98	248	28.33

[a]Brown rot infected fruits distributed among sound lemons in field boxes 29 January, 1961; inspections for contact infection 6 February, 1961, and 9 February, 1961.

comparison with those treated. In one treatment the lemons were immersed for 4 minutes in a solution of 2.5% soda ash - 0.5% soap, the initial temperature of which was 49°C (120°F). At the end of 4 minutes the solution had cooled to 47°C (116°F). The fruits then received a 3-second sprinkling with water and a 5-second sprinkling with water wax emulsion[1]. In the other treatment the lemons were immersed for 3 minutes in a 0.375% solution of sodium-ortho-phenylphenate plus detergent (pH 12.0) which had an initial temperature of 49°C (120°F) and cooled to 47°C (117°F) by the end of the treatment. Sprinklings with water and water wax emulsion followed as in the first treatment.

After being held at laboratory temperature at 22°-27°C (72°-80°F) for 3 to 8 days the lemons were examined for contact decay (Table 4).

Table 4 shows that at 20°C (68°F) 13.9% of the treated and 28.33% of the untreated lemons decayed, while at 10°C (50°F) only 0.6% of the treated and 11.18% of the untreated fruits decayed. This would seem to indicate the desirability of holding the fruits at low temperatures prior to washing and the hot solution treatment. However, cold turgid fruits fresh from the orchard are injured by immersion in the hot solution; rind oil is liberated, causing a rind breakdown called oleocellosis. To avoid this injury the treatment must be delayed until the rind surface is slightly dried. As an added precaution a strong soap solution (0.5%) is used in the washing tank to emulsify any rind oil that may be released and thus prevent its corroding the fruit surface.

The hot solution treatments showed little difference in effectiveness. One thousand lemons treated with soda ash soap for 4 minutes had a total of 62 decays (6.2%) while those treated with sodium-ortho-phenylphenate had 72 decays (7.2%). This slight difference can be accounted for by the additional minute immersion in the soda ash-soap solution. So far as brown rot control in the packinghouse is concerned, hot water without chemicals would be just as effective as the solutions. The chemicals are added for cleansing and for control of rots caused by blue-green molds and other fungi.

[1]Number 22 water wax emulsion made at Sunkist Field Department Laboratory; 0.75% used.

Literature Cited

1. KLOTZ, L. J. 1941. Brown rot of citrus fruit. Important factors in its control in orchard and packing house. California Citrog. 27: 6, 23.
2. KLOTZ, L. J. 1943. Phytophthora infections of citrus and their control. California Citrog. 28: 200-201, 220-221.
3. KLOTZ, L. J., and T. A. DeWOLFE. 1960. The production and use of zoospore suspensions of Phytophthora spp. for investigations on diseases of citrus. Plant Disease Reptr. 44: 572-573.
4. KLOTZ, L. J., and T. A. DeWOLFE. 1961. Limitations of the hot water immersion treatment for the control of Phytophthora brown rot of lemons. Plant Disease Reptr. 45: 264-267.
5. KLOTZ, L. J., and H. S. FAWCETT. 1941. Brown rot and gummosis. California Citrog. 26: 114, 142.

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REACTION OF TOMATO VARIETIES AND BREEDING LINES TO FUSARIUM OXYSPORUM F. LYCOPERSICI RACE 1

Warren R. Henderson and N. N. Winstead

Summary

Of the 100 entries tested for resistance to Fusarium oxysporum f. lycopersici race 1, 55 were resistant, 43 susceptible, and 2 segregated for resistance. Thirty-nine S. T. E. P. (Southern Tomato Exchange Program) breeding lines were tested and 34 were resistant and 5 susceptible. Each of the varieties or lines classed as resistant is considered to possess as a part of its genetic complement type A (genotype - Ii or II) resistance.

The tomato breeder transferring monogenic resistance to Fusarium wilt from a resistant to a susceptible tomato variety need no longer transmit, in conjunction with resistance, all of the many undesirable horticultural characters found in the original source of resistance, Lycopersicon pimpinellifolium accession 160. Since the release of the Pan America tomato variety (5), numerous Fusarium wilt resistant varieties have been developed. Reports on the release of varieties are widely scattered in various technical and miscellaneous publications, and do not always indicate whether the variety carries type A (monogenic) or type B (multigenic) resistance. Consequently, a single current reference, rather than the many isolated references now available of the reaction of many of the important varieties of Lycopersicon esculentum to Fusarium wilt, should prove an aid to the tomato breeder. The reaction of 100 tomato varieties and breeding lines to race 1 (4) of Fusarium oxysporum f. lycopersici (Sacc.) Snyder & Hansen is reported here.

MATERIALS AND METHODS

Seed was sown in steam disinfected sand. At the expanded cotyledon stage, the seedlings were transplanted to flats containing a steam disinfected soil-peat moss mixture. Each flat contained 35 plants of the variety to be tested, 7 plants of the susceptible check Rutgers, and 7 of the resistant check variety Homestead 24, or a population of 49 plants per flat.

Inoculum was prepared using a highly pathogenic isolate (R5-6) of the tomato wilt organism. Cultures were grown in 100 ml of a modified Richard's solution (7) in 16-ounce pharmaceutical bottles placed on the side to give the greatest possible exposure to air. The culture fluids were filtered off in a Büchner funnel and discarded, and the mycelial mats placed in tap water and macerated in a blender. The mycelium from one bottle of culture was used to inoculate each flat of tomato plants.

Prior to inoculation, greenhouse air temperature was maintained at about 70° to 75° F day, and 60° to 65° night, the lower day and night temperatures were maintained during periods of cloudy weather. The roots were severed by running a sharp knife alongside each row of plants and the inoculum poured (50 ml of mycelial suspension per row) in the furrow when the plants were at the 4-leaf stage (approximately 3 weeks following transplanting). Greenhouse air temperature was then raised to 75° to 85°, both day and night. Soil moisture was maintained at a level which promoted relatively rapid vegetative growth. A second inoculation was made 10 days following the first. The seedlings were assayed 30 days from the initial inoculation. Plants were placed in 1 of 4 categories according to visual symptoms: 1 - healthy; 2 - no wilting but slight to moderate vascular discoloration; 3 - distinct wilting and vascular discoloration; and 4 - severe wilting or dead. Plants in classes 2, 3, and 4 were considered susceptible, and those in class 1 resistant.

RESULTS AND CONCLUSIONS

The reactions of the tomato varieties and breeding lines to inoculation with F. oxysporum f. lycopersici race 1 were as follows: a) Resistant: Campbell 146, FC7-WF3-B, FC7-WG3-B, Homestead F-M, Homestead 24, Indian River, Kokomo, Kopiah, Manalee, Manalucie, Manasota, Marion, N. C. 201, Ohio WR Jubilee, Pa. 117, Pearson VF 6, S. B. W. 4i-1, S. B. W. 6i-2, Texto 2, U. S. 357, #670, S. T. E. P. 275, 281, 284, 287, 305, 311, 314, 324, 326, 327, 341, 344, 345, 346, 347, 348, 349, 350, 351, 352, 353, 354, 355, 356, 357, 358, 359, 360, 361,

362, 363, 365, 366, 369; b) Susceptible: Burpee's Big Boy Giant Hybrid, Burpee's Big Early Hybrid, Burpee's Sunnybrook Earliana, Burpeeana Early Hybrid, Cardinal Hybrid, Cluster-Mato, Cooper's Special x M84, C. R. T. Hybrid, Cuyano, Earliana, Early Delicious Hybrid, Early Giant Hybrid, Early Prolific Hybrid, Fireball, Fordhook Hybrid, German tomato, Glamour, Marglobe, Marman, Marmande, Mendoza 44, Moreton Hybrid, New Glecano, New Wonder Boy Hybrid, Pa. 103, Pearson S, Peron, Plamar,* Pritchard, Queens, Rhode Island Early, Rutgers, Sioux, Thessaloniki, Valiant, Vancross, 30-A-1, 41-A-1, S.T.E.P. 322, 329, 330, 367, and 368.

A comparison can be made of the response of each variety and line with that of the corresponding susceptible and resistant check variety planted in the same flat[1]. An occasional escape was seen in the susceptible check and in a presumably susceptible variety. Environmental induced "resistance" in type B resistant plants could be responsible (3, 6). A more rapid soil moisture loss undoubtedly occurred at the edge of those flats at the sides of the bench. Plants in this area would have a greater carbohydrate reserve resulting from a more limited vegetative growth. Less root damage also possibly occurred during cutting due to a restricted root area. Differences in the quantity of inoculum could also have been a contributing factor. Even if none of the aforementioned factors were responsible, the number of escapes was small and probably of little consequence.

Most of the susceptible plants fell into either class 3 or 4. The inoculation procedures were quite severe and were designed to eliminate plants which carried only type B (multigenic) resistance. For example, the susceptible check Rutgers carries type B resistance and only 11 plants out of more than 500 plants inoculated were classed as resistant, conversely only 1 out of more than 500 Homestead 24 (type A resistant check) plants was killed. The low level of resistance of type B resistant varieties prevents their detection in this test.

As expected in an F_2 hybrid, Stokescross #6 segregated approximately 3 resistant: 1 susceptible plant. The line designated C. P. 1627 presumably was a backcross progeny of a heterozygous parent (Ii) to a susceptible parent (ii), since the line segregated in a 1:1 manner.

The high level of resistance found originally by Bohn and Tucker (2) in Lycopersicon pimpinellifolium, Accession 160, has obviously led to a concentrated effort by the tomato breeder to incorporate this single dominant gene for resistance in many of the current and new varieties. Accession 160 was found by Alexander and Tucker (1) to segregate for resistance when inoculated with a second race of the Fusarium wilt fungus found in Ohio. In addition to the single dominant gene imparting resistance, Alexander and Tucker postulated that complimentary genes were required in addition, to produce resistance to race 2. Fortunately, from the commercial viewpoint, race 2 remains limited in its distribution. Thus far race 2 resistance has received little consideration in breeding for Fusarium wilt resistance.

Literature Cited

1. ALEXANDER, L. J., and C. M. TUCKER. 1945. Physiologic specialization in the tomato wilt fungus Fusarium oxysporum f. lycopersici. J. Agr. Research 70: 303-313.
2. BOHN, G. W., and C. M. TUCKER. 1940. Studies on Fusarium wilt of the tomato. I. Immunity in Lycopersicon pimpinellifolium Mill. and its inheritance in hybrids. Missouri Agr. Exp. Sta. Research Bull. 311: 82 pp.
3. FOSTER, R. E., and J. C. WALKER. 1947. Predisposition of tomato to Fusarium wilt. J. Agr. Research 74: 165-185.
4. GERDEMANN, J. W., and ARTHUR M. FINLEY. 1951. The pathogenicity of races 1 and 2 of Fusarium oxysporum f. lycopersici. Phytopathology 41: 238-244.
5. PORTE, W. S., and H. B. WALKER. 1941. The Pan America tomato, a new red variety highly resistant to Fusarium wilt. U. S. Dept. Agr. Circ. 611.
6. WALKER, J. C., and R. E. FOSTER. 1946. Plant nutrition in relation to disease development. III. Fusarium wilt of tomato. Am. J. Botany 33: 259-264.
7. WINSTEAD, N. N., and J. C. WALKER. 1954. Production of vascular browning by metabolites from several pathogens. Phytopathology 44: 153-158.

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[1]Detailed data for each variety and line as well as a list of seed sources will be supplied by the author in mimeograph form upon request by the reader.

PHRAGMIDIUM ROSE RUST EPIDEMIC IN LOUISIANA STATE UNIVERSITY GARDENS

I. L. Forbes and T. P. Pirone

In mid-January of 1961 rust was observed on leaves of rose bushes in the Horticulture and Entomology Rose Gardens on the Louisiana State University Campus. Most plants were still in full leaf, despite winter frosts and ice, and both uredia and telia occurred on leaves only. A careful survey revealed that the disease had attained epidemic proportions. Of the 117 varieties examined in the garden, all but five showed rust. However, there was a great range in the amount and severity of rust on different varieties. In Table 1 the varieties are given a susceptibility rating, based on the severity of rust infection on each.

Table 1. Varietal reaction to rust.

No rust	Severe rust infection		Light rust infection	Very light infection
Lionel Barrymore	Ruby Lips	47-3942	Heat Wave	Fashionette
554	Ardelle	Siren	Green Fire	Amy Vander-
Starlet	Kordes Perfecta	Ma Perkins	513	bilt
Greenrose	Arlene Francis	Vogue	Maxine	Gold Cup
Summer Snow	Pink Frost	Confidence	Isobel Harkness	The Fair
	Reno	Chief Seattle	Fanfare	54-14811
	Betsy McCall	Helen Traubel	Firecracker	Living
	Spartan	White Swan	2455	Red Radiance
	Fusilier	New Yorker	Midnight	Etiole De Hol-
	49-1182	Big Daddy	Aztec	lande
	49-28	Gay Lady	5706	Peace
	Jeanie	Buccaneer	47-22031	Lilibet
	Audy Murphy	Chrysler Im-	Gladiator	Garden Party
	White Knight	perial	4471	Connie Mac
	5014-55	Embers	Fort Knox	Hedda Hopper
	4342-2	Frolic	White Bouquet	Fred Howard
	Konrad Adenauer	Bravo	Sun Valley	Snow Bird
	Montezuma	Pink Chiffon	Queen Elizabeth	Mrs. E. P.
	Better Times	Sweet Sixteen	Wild Fire	Thom
	53-9833	Ondine	Circus	Applause
	54-8881	Grand Duchess	47-18201	Girona
	Pink Peace	Charlotte	Golden Girl	Debonair
	56R52C-P	Heart's Desire	47-889	
	Golden Masterpiece	Christopher	Pink Radiance	
	Jiminy Cricket	Stone	Talisman	
	Baby Blaze	Mrs. Sam Mc-	56R51C-P	
	Golden Rapture	Greedy	Taffeta	
	Nocturne	McGredy Ivory	Rose of Freedom	
	Virgo	Sutter's Gold		
	Rubiuiat	Mme. Chiang Kai-		
	The Doctor	shek		
	White Briarcliff	Mary Margaret		
	McGreedy's Yellow	McBride		
	Suzon Lotthe	Roundelay		
	Redcap	La Jolla		

In 1950 Plakidas published a record[1] of rust on a rose specimen from Newellton, Louisiana. The Newellton rose bushes had been obtained from California. This was the first record of the rust in Louisiana. The writers were informed by Drs. Hanchey and Kimbrough of the Horticulture Department that rust had been observed in the Louisiana State University Horticulture Rose Garden for several years prior to 1961.

A limited survey of home garden roses in the Baton Rouge area, by Pirone and Forbes in January 1961, did not reveal rust on rose plants. However, Dr. John Roussel informed us that he had seen rust on plants in his home garden and he surmised that he had inadvertently transferred the inoculum from his Entomology plots near the Louisiana State University Campus. Drs. Roussel, Hanchey and Kimbrough told us they had obtained rose bushes from California in past years for experimental plantings.

Since the rust has been known to occur in California for a number of years, it is probable that the disease was introduced into the Louisiana State University gardens, as well as into the North Louisiana (Newellton) area, from California

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[1] Plakidas, A. G. 1950. Rose rust (Phragmidium americanum ?) in Louisiana. Plant Disease Reptr. 34:197.

IN-THE-FURROW APPLICATION OF SOIL FUNGICIDES FOR CONTROL OF COTTON SEEDLING DISEASES[1]

Charles R. Maier[2]

Abstract

Evaluations were conducted on the effectiveness of in-the-furrow application of soil fungicides and methods of applying them to control cotton seedling diseases. From a total of 17 chemicals and chemical combinations evaluated in field trials, the most effective were PCNB, PCNB+captan, captan+Phaltan, and thiram. Application of the materials evaluated in the form of low-volume sprays appeared to be more effective than in-the-furrow dusts or planter-box dusts. The cost-return considerations of the results of 2 years' testing indicate that the use of in-the-furrow fungicides is practical, and that returns will range from six to ten times the application costs. In-the-furrow application of chemicals resulted in disease loss reduction, stand and yield increases, earlier maturity, and generally more vigorous and healthy plants than the untreated checks. The tests were conducted on irrigated cotton of different varieties and under varying growing conditions over New Mexico.

The seedling disease complex in New Mexico causes an annual loss of 2 to 2 1/2% of the potential cotton crop. In 1959 this loss was estimated at $1,350,000 (4). Over the entire cotton belt of the United States, losses approach 2 1/2%. The seedling disease complex is composed of several soil-inhabiting fungi, including *Rhizoctonia solani*, *Thielaviopsis basicola*, *Fusarium* spp., and in some areas *Pythium* spp. The latter organism does not appear to be a factor in New Mexico.

Seedling disease injury takes several forms, which include seed rot, pre-emergence damping-off, post-emergence damping-off, seedling root-rot, and soreshin. The seed-rot phase has been largely overcome by the widespread use of high quality seed, treated with suitable seed treatment chemicals. Recent research in Texas (7), Arizona (4), and other areas has demonstrated seed treatment to be inadequate in controlling other phases of seedling diseases. Once the seedcoat has been ruptured during germination, the young seedlings have no protection from pathogenic soil fungi. Attempts have been made to afford protection to cotton seedlings by mixing soil fungicides with the covering soil at the time of planting (3), and have been demonstrated to be an effective measure for controlling seedling disease fungi (2). The measure is now recommended for control of the seedling complex in Texas (3).

In-the-furrow application of fungicidal materials has been studied in New Mexico since the early 50's, and has given indication of practical control. The lack of effective control reported by Wiedman (1) in 1957 and 1958 was attributed to an extremely light incidence of seedling disease in the experimental fields. The current investigations were carried out in the Mesilla Valley in 1959 on five farms in grower-cooperative experiments, and in six locations widely spaced over the State in 1960. The methods of applying fungicides described by Ranney and Hillis (8) were adapted to these experiments. Low-volume sprays, dusts, and planter-box dusts were utilized.

RESULTS OF IN-THE-FURROW FUNGICIDE TRIALS

In-the-furrow fungicide trials were conducted on five fields in the Mesilla Valley in 1959. Chemicals in fields 1, 2, and 3 were applied as low-volume sprays at 50 gallons/acre, and in fields 4 and 5 as dusts. Sixteen materials were tested in randomized block design with 8 or 10 treatments in each field. Four replications of 4-row plots ranging from 560 to 940 feet in length were used, and water check plots as well as untreated check plots were included in all spray experiments. The experimental fields were chosen for a history of severe seedling disease incidence. The incidence of post-emergence damping-off in the plots ranged from 22% to 40% of emerged plants. Data taken from the two center rows in each 4-row plot included stand

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and disease incidence 3 to 4 weeks after planting and yield in seed cotton per plot or yield in seed cotton estimated from boll and stand counts in September. The results of in-furrow spray trials are presented in Table 1, and of in-furrow dust trials in Table 2.

Table 1. Results of 1959 in-the-furrow spray application of soil fungicides for control of cotton seedling diseases[a].

Material[c] and rate (pounds/acre)	Post-emergence loss (% of emergence)			Stand 3-4 weeks after planting (seedlings/10 feet)			Lint cotton yield[b] (pounds/acre)		
	Field			Field			Field		
	1	2	3	1	2	3	1	2	3
Captan + Phaltan 2+2	9	13	14	69	87	64	2043	1547	1420
PCNB + captan 2+2	14	10	-	59	72	-	1716	1623	-
Shell 4741, 4	16	13	12	69	88	57	1794	1584	1200
Panogen SD, 4	14	8	12	60	78	63	1688	1620	1120
Maneb (Dithane M-22), 5	11	-	12	66	-	56	1925	-	1080
Zineb (Dithane Z-78), 5	14	-	16	61	-	58	1674	-	1115
Nabam (Dithane D-14), 4	20	18	15	57	67	61	1588	1599	985
Nabac 25, 5	16	-	-	61	-	-	1792	-	-
Zineb + captan 3+2	-	16	-	-	68	-	-	1593	-
Ferbam, 5	-	-	16	-	-	58	-	-	940
Nabac 25 + maneb 3+2	-	10	-	-	74	-	-	1620	-
Water check	24	20	28	49	56	51	1424	1322	835
Untreated check	30	22	40	45	50	49	1268	1176	745
LSD .01	12	10	18	14	13	9	164	146	136

[a]Materials applied in 50 gallons/acre, water check 50 gallons/acre.
[b]Lint cotton yields calculated from seed cotton weights per plot except field 3, which were estimated from boll counts taken Sept. 12, 1959.
[c]captan = N-trichloromethylmercapto-4-cyclohexene-1,2-dicarboximide; Phaltan = N-trichloromethyl thiophthalimide; PCNB = pentachloronitrobenzene; Shell 4741 = O,O,O-trimethylphosphorothioate; Panogen SD = methylmercury hydroxide 3.5%; maneb = manganese ethylene bisdithiocarbamate; zineb = zinc ethylene bisdithiocarbamate; nabam = disodium ethylene bisdithiocarbamate; Nabac 25 = 2' methylenebis (3,4,6 trichlorophenyl); ferbam = ferric dimethyl dithiocarbamate.

In all trials, at least two materials were superior to the others included, which were: Field 1, captan + Phaltan and maneb; Field 2, PCNB + captan, Nabac 25 + maneb, and Panogen SD; Field 3, captan + Phaltan and Shell 4741; Field 4, thiram (Tersan 75) and captan + Phaltan; and Field 5, Phaltan + zineb and captan + Phaltan. All materials in the trials significantly reduced seedling disease incidence except nabam in Field 1 and maneb in Field 4. Yield increases were generally associated with stand increases and somewhat less with disease control. The notable exception was Phaltan + zineb in Field 5, which increased yield but showed no significant stand increase.

From the field and greenhouse trials of 1959, four materials were selected for evaluation of effectiveness in disease control: PCNB, PCNB + captan, captan + Phaltan, and thiram. Five fields were selected for the 1960 trials in different cotton-growing areas of the State, and all experiments were conducted with the four materials applied as both low-volume sprays and planter-box dusts. A water check and an untreated check were also included in each field. The locations were: Plot 1, Portales; Plot 2, Carlsbad; Plot 3, Roswell; Plot 4, Las Cruces; and Plot 5, Deming. The objective of the trials was to evaluate the effectiveness of the chemicals as well as the application methods under widely varying growing conditions. Each experiment contained four replications of 4-row plots ranging from 650 to 1280 feet in length. Seedling rates varied from 20 to 26 pounds/acre and the plots were planted between April 20 and May 2, 1960. The spray materials were applied in-the-furrow in 25 gallons spray/acre, and the dry materials were mixed with the seed in the planter-box. The results of the 1960 in-furrow application trials are given in Table 3. Data were taken from the middle two rows of each plot.

From all trials, two conclusions may be made: 1) PCNB alone is superior to other materials for controlling cotton seedling diseases; and 2) spray application is a more efficient method of applying soil fungicides for seedling disease control than is planter-box application. With relatively few exceptions, each material performed better in giving stand and yield in-

Table 2. Results of 1959 in-the-furrow dust application of soil fungicides for control of cotton seedling diseases[a].

Material and rate (pounds/acre)	Post-emergence loss (% of emergence) Field 4	Post-emergence loss (% of emergence) Field 5	Stand 3-4 weeks after planting (seedlings/10 feet) Field 4	Stand 3-4 weeks after planting (seedlings/10 feet) Field 5	Lint cotton yield[b] (pounds/acre) Field 4	Lint cotton yield[b] (pounds/acre) Field 5
Captan + Phaltan 2+2	8	6	60	61	1275	1312
PCNB + captan 2+2	10	8	56	49	1075	1230
Maneb (Dithane M-22), 5	20	-	53	-	1035	-
Zineb (Dithane Z-78), 5	12	-	55	-	1150	-
Nabac 25, 5	12	-	59	-	1230	-
Zineb + captan 3+2	-	10	-	48	-	1288
Ferbam, 5	14	-	53	-	1050	-
Chloranil[c] (Spergon), 5	11	16	55	50	1020	1266
Thiram[c] (Tersan 75), 5	9	12	60	59	1295	1244
Dichlone[c] (Phygon), 5	12	-	54	-	1005	-
Nabac 25 + captan 3+2	-	16	-	50	-	1262
Phaltan + zineb 3+2	-	14	-	56	-	1369
Untreated check	27	29	44	48	1090	1174
LSD .01	9	12	14	10	92	120

[a]Wettable powder or dust formulation applied in the furrow at planting.
[b]Lint cotton yield field 4 estimated from boll counts taken Sept. 15, 1959 and field 5 calculated from weights of seed cotton per plot.
[c]chloranil = tetrachloro-p-benzoquinone; thiram = tetramethylthiuram disulfide; dichlone = 2,3-dichloro-1,4-naphthoquinone.

creases when applied as sprays, and the magnitude of the increases were considerably greater, than when applied via the hopper box. This comparison is shown in Table 4, as the mean performance of the materials at all five locations.

Several more or less intangible or visual factors were observed in the evaluations of both materials and application methods. In spray-treated plants, a greater percentage of the bolls were mature than in dust-treated plants or checks when boll counts were taken on October 1. Seedling root development was more extensive, plant growth was better, and seedlings were more vigorous in spray-treated plots than either dust-treated plots or checks when data was taken after 3 to 4 weeks. The percentage of plants infected was lower and the injury to infected plants less severe in the spray-treated plants than other treatments. These parameters were compared by the examination of 25-plant samples collected at random from each plot approximately 4 weeks after planting.

DISCUSSION AND CONCLUSIONS

In consideration of suitable application methods, the use of a low-volume spray is preferred by the author to either mechanical dusting or planter-box application. The first reason is in the greater magnitude of increases obtained in the same fields where the same materials were compared, and measured both in disease control and lint yield increase.

A second reason is that where soil moisture is low to fair, the application of 25 gallons of spray/acre, half of which is directed into the knife furrow onto the seed, will supply immediate moisture for rapid germination. When dust formulations are applied, necessitating the complete wetting of the dry material before sufficient moisture reaches the seed, germination may be considerably delayed where moisture is low. In one location (Plot 3, Roswell) visual growth differences estimated at 2 to 4 days by the end of 3 weeks after emergence were apparent; the moisture level was considered good by the cooperator at planting. In order to obtain full benefit from in-furrow dust application, it appears that soil should be moderately moist at planting and treatment. At another location (Plot 2, Carlsbad) this condition occurred, and growth differences, while present, were slight.

A third factor favoring in-furrow spray application of soil fungicides over in-furrow or planter-box dust application lies in the earlier maturity of bolls in the spray-treated plots. At two locations in the 1959 trials (Fields 1 and 3) a mean of 56% of total seed cotton in spray-

Table 3. 1960 response of cotton treated for seedling disease control with four materials applied as in-the-furrow sprays and planter-box dusts.

Material	Rate tech. (pounds/acre)	Method	Disease loss (% of emergence) Field[a] 1	2	3	4	5	Stand 3-4 weeks (seedlings/10 feet) Field 1	2	3	4	5	Lint cotton yield[b] (pounds/acre) Field 1	2	3	4	5
PCNB (Terraclor)	3.5	spray	10	8	6	7	8	54	68	45	76	56	684	1373	1181	1220	623
PCNB+ captan	2 + 2	spray	13	11	9	10	10	49	58	39	68	53	652	1258	1025	1097	560
Captan + Phaltan	2 + 2	spray	12	10	8	9	10	48	66	40	70	54	655	1262	1094	1134	614
Thiram (Arasan)	3.5	spray	13	11	10	11	10	46	58	38	65	52	636	1196	1012	1000	537
Water check	25 gal.	spray	15	16	12	14	14	41	51	33	60	46	565	1099	909	925	482
PCNB	3.5	dust	12	10	10	11	11	44	57	37	60	49	589	1153	910	1005	528
PCNB + captan	2 + 2	dust	13	11	11	10	12	44	54	35	58	48	568	1081	910	1001	508
Captan + Phaltan	2 + 2	dust	12	10	11	10	9	45	55	37	58	49	572	1134	980	1061	521
Thiram (Arasan)	3.5	dust	13	13	11	11	13	43	53	34	56	46	561	1103	929	1014	503
Untreated check		--	18	20	15	16	16	38	46	31	53	40	542	1030	841	930	486
LSD .01			2.2	4.1	2.8	2.3	3.2	5	8	7	10	6	45	98	84	100	64

[a] Fields located: 1, Portales; 2, Carlsbad; 3, Roswell; 4, Las Cruces; and 5, Deming. Materials applied in 25 gallons water/acre and dry wettable powders at each location. Stand and disease loss data were collected 3 to 4 weeks after each planting by random stand counts.

[b] Yields in lint cotton/acre calculated from stand and boll counts taken at random from middle two rows of each plot Sept. 29 to Oct. 8, 1960.

treated plots was collected at the first picking, compared with 45% for the check. Early maturity of cotton in the spray-treated plots was observed at one location in the 1960 trials as well (Field 5, Deming). This earlier maturity would appear to give additional return to the grower through higher quality lint and result from more uniform stands where escape from infection had been obtained from better control of seedling disease fungi as well as where better early growth of seedlings had occurred.

In any consideration of disease control with chemicals, the practical economics must be dealt with. That in-the-furrow application of fungicides is both effective and profitable is reported by Porter, Bird, and Smith in Texas (7) and supported by Maier (6) in New Mexico. From the costs of chemicals applied in the 1959 trials and the results obtained, increases of considerably greater magnitude than reported above were obtained. For the spray application of effective chemicals, material outlays from $5 to $12 per acre produced returns of $60 to $130 per acre. It should be pointed out, however, that all experiments were conducted under almost ideal conditions to show this magnitude of increased return -- a moderate to high seeding rate, soil conditions favoring infection of seedlings, and a high prevalence of seedling disease -- while in practice, returns may be of a much lower order. The increased return would be dependent upon seedling disease severity in each specific location. Where the mean disease loss in the experimental plots was 29.6% of emerged plants, the estimated mean stand loss for the State was 11.6% (5). Even with this consideration, the in-furrow application of soil fungicides for seedling disease control appears to be a paying proposition. This is borne out in the 1960 trials, which gave a more direct evaluation of each chemical as both in-furrow spray and planter-box dust in a variety of conditions. Returns estimated at $25 to $60 per acre were obtained where the application costs ranged from $5 to $9 per acre for material.

Table 4. Comparison of the effectiveness of soil fungicides applied as in-the-furrow spray and planter-box dust for control of cotton seedling diseases, as means of five 1960 cooperative trials.

Material	% disease loss to stand	Lint yield (pounds/acre)	Lint increase over check[a] (pounds/acre)	Increased cash return/acre[b] over dust	Increased cash return/acre[b] over check[a]
PCNB (Terraclor)	7.8	1016.2	220.2	$59.13	$74.87
PCNB + captan	10.6	918.4	122.4	34.58	40.39
Captan + Phaltan	9.8	951.8	155.8	32.40	51.41
Thiram (Arasan)	11.0	876.2	80.2	17.89	26.47
Check (water)	14.2	796.0	--	--	--
PCNB	11.2	837.0	71.2	--	23.50
PCNB + captan	11.4	813.6	47.8	--	15.77
Captan + Phaltan	10.4	853.6	87.8	--	28.97
Thiram (Arasan)	12.2	822.0	56.2	--	18.55
Untreated check	17.0	765.8	--	--	--

[a]Lint yield increase and cash return for spray treatments compared with water check; dust treatments compared with untreated check.
[b]Based on 33¢ per pound for lint.

Literature Cited

1. ANONYMOUS. 1958. Seedling diseases of cotton. 1957 Annual Report, Dept. of Botany and Entomology, (Unpublished), New Mexico Agr. Exp. Sta., University Park. pp. 97, 104.
2. BIRD, L. S. and C. D. RANNEY. 1958. Fungicides mixed with covering soil at planting for cotton seedling disease control. Texas Agr. Exp. Sta. Progress Report 2001. pp. 1-3.
3. BIRD, L. S., C. D. RANNEY, and G. M. WATKINS. 1957. Evaluation of fungicides mixed with the covering-soil at planting as a control measure for the cotton seedling-disease complex. Plant Disease Reptr. 41: 165-173.
4. CHILTON, J. E. 1960. Cotton seedling diseases. Crop Comments, Arizona Chem. and Fertilizer Co., Phoenix. (April) 14(6): 1-4.
5. MAIER, C. R. 1959. Survey of cotton seedling disease prevalence and severity of losses in New Mexico in 1959. Plant Disease Reptr. 43: 1048-1049.
6. MAIER, C. R. 1960. Current research in cotton seedling disease control by chemical means. Address, Proc. 3rd New Mexico Agr. Chem. Conf., University Park. Jan. 13, 1960. pp. 6-10.
7. PORTER, D. D., L. S. BIRD, and N. B. SMITH. 1959. Value of cotton seedling disease control. Plant Disease Reptr. Suppl. 259: 205-208.
8. RANNEY, C. D., and A. M. HILLIS. 1958. Methods of applying fungicides in the covering soil at planting for controlling seedling diseases of cotton. Texas Agr. Exp. Sta. Progress Report 2001. pp. 1-3.

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ACHAPARRAMIENTO (CORN STUNT)

Oscar Ancalmo and William C. Davis[1]

Achaparramiento, the corn (Zea mays) stunt virus disease, was first recorded in El Salvador in 1959 on a farm in the Pacific coastal area.

During the principal corn-growing months (May-August) of 1960 a survey showed that the disease was generally established in fields along and near the south coast. The survey further indicated that hybrid, commercial open pollinated varieties, and self pollinated lines of corn were all attacked by the disease. Some individual field studies on H-501 and H-503, two of the more widely grown hybrids in this country, showed above 75% of the plants diseased and yield reductions of as much as 53% in some cases.

The Rio Grande and Mesa Central types of the disease described by Maramorosch[2] for Mexico have been observed in El Salvador with little variation. Apparently the Mesa Central type in El Salvador shows a lesser degree of stunting than in Mexico; the Rio Grande type shows a more severe degree of stunting. A mixture of both types not previously reported has been found in El Salvador. Here, the plants may show the typical symptoms of the Mesa Central type on the lower portion of the plant and that of the Rio Grande type in the upper portion or vice versa. Plants having this combination of symptoms are moderately stunted.

What appears to be a third type of the disease has also been observed by the authors. Diseased leaves show a fine stipple striping along the blades. These small chlorotic spots have not been observed to form bands but stay separate. Such plants are not stunted. This type of the disease has been transmitted in screened cages by the insect Dalbulus maidis (Del. & W.) Large populations of this insect were noted in all fields where any type of the disease was observed.

Sweet corn apparently is more susceptible than commercial field corn. In some instances 100% infection was recorded, causing a complete loss of yields.

A search for disease resistant corn material has already been initiated at the Dirección General de Investigaciones Agronómicas. Local material together with material sent by the Rockefeller Foundation program in Mexico is being tested. Preliminary field data indicate the existence of a resistant factor.

DIRECCIÓN GENERAL DE INVESTIGACIONES AGRONÓMICAS AND UNITED STATES OPERATIONS MISSION, SANTA TECLA, EL SALVADOR, C. A.

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[2]Maramorosch, Karl. 1955. The occurrence of two distinct types of corn stunt in Mexico. Plant Disease Reptr. 39: 896-898.

OILS REDUCE SPORULATION OF SEPTORIA ON CELERY

J. D. Wilson[1]

Abstract

Petroleum oils, such as those used for 75 years to control certain insects, have not come into use to control plant diseases since they are not really fungicides, and many of them, especially those with high viscosity ratings, are injurious to plant foliage. However, various oils do act in some manner to control certain diseases, apparently by disturbing the usual host-parasite relationship after infection has occurred. Unfortunately, from the use standpoint, the lighter oils, with viscosity ratings below 65 to 70 Saybolt Universal Seconds (SUS), which are least phytotoxic, are also least effective in disease control. Also, plants vary widely in oil tolerance, with celery and beet less sensitive than tomato and potato. Certain oils were found to greatly reduce the formation of pycnidia in the lesions of late blight on celery (*Septoria apii*), although they were comparatively ineffective in preventing initial infection by the fungus. This acted in turn to reduce the degree of secondary infection, and thus to partially check the further advance of the disease and to increase the yield over that of the untreated check plants. Different fungicides responded differently to formulation with oil, with Tribasic and ziram being increased in effectiveness in blight control more noticeably than maneb or Dyrene.

Petroleum oils have been used for 75 years to control such pests as aphids, scale insects, and mites, but have seldom been used in the control of fungus diseases, chiefly because they are not fungicides in the true sense of the word and because most of them are injurious to plant foliage. Furthermore, various investigators have found that the oils that have been most effective in checking disease development were also most likely to be phytotoxic to the host plant.

Since oils as a group do not interfere with germination of fungus spores on the surface of the leaf, or subsequent penetration of the leaf tissue to cause infection, it seems likely that they must act later in some way to disturb the usual host-parasite relationship in such a way that the fungus is not able to develop and reproduce as vigorously as it might otherwise do.

Several oils, differing chiefly in their viscosity ratings (a value which is usually stated as so many Saybolt Universal Seconds (SUS) at 100° F) have been applied alone, and with various fungicides, to potato, tomato, cucumber, carrot, celery, and sugar beet during the past 3 or 4 years in Ohio in an effort to determine their effect on disease control. The results have been quite varied, depending upon the oil used, the plant to which it was applied, and the disease in question. Also, the fungicides have been found to be affected differently by the addition of oil to the spray formulation. Of the crop plants listed above, the tomato is most subject to oil injury. Although some decrease in disease activity has been noted in specific instances, it seems unlikely that any of the oils now available can be used successfully, either alone or as fungicide supplements, to control tomato diseases. This is true largely because the injurious effects of the oil on growth and yield more than offset any benefit that may be derived from disease control Potatoes are more oil-tolerant than tomatoes, but here again any improvements in foliage condition or yield have not been significant. Cucurbits such as muskmelon and cucumber are tolerant of small quantities of certain oils, and in some instances the use of oil as a supplement to certain fungicies has aided in the control of powdery mildew.

Carrot, celery, and sugar beet are all subject to infection by *Cercospora* species and since certain petroleum oils have given control of the Sigatoka leaf spot of banana, the causal organism of which is also a *Cercospora*, it was thought worthwhile to investigate the possibility that the Cercospora leaf spots of these crop plants could also be controlled with oil. There has been some evidence of such control, but infection in sufficient degree to cause any appreciable reduction in yield has not been present in any of the experimental plots, and thus it has not been possible to obtain any definite control data. All three crops have been found comparatively tolerant to oil injury.

It was planned in the beginning of this study on the control of vegetable diseases by the use of oil to make a special effort to control Cercospora leaf spot (early blight) of celery. How-

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ever, the disease has not occurred in epidemic form in the experimental area during any one of the last 3 years. Instead late blight, caused by Septoria apii, has been severe enough each year to render the plants in the untreated check plots unfit for sale, and thus it is to the effect of oil, applied alone or in combination with various fungicides, on this disease, that the data presented here will be restricted.

In 1958 three oils were applied to celery at the rate of 1.5 gallons/acre/application at the Muck Crops Substation, and one of these oils was also added to each of five different fungicides: manganese ethylene bisdithiocarbamate (maneb) (Manzate); zinc dimethyl dithiocarbamate (ziram) (Zerlate); N-trichloromethyl thiophthalimide (Phaltan); 2,4-dichloro-6-(o-chloroanilino) -s-triazine (Dyrene); and Tribasic. The fungicides were also applied without the addition of oil. Soybean was the source of one of the oils used and the other two were petroleum oils; namely, Sun Spray Oil No. 7 and ML-100A (which is a mixture of two different oils). The soybean oil had little effect on disease or on yield. The two petroleum oils did give a considerable reduction in the defoliation caused by late blight, and a yield increase over the untreated check plots. The Sun Spray Oil No. 7, with a viscosity rating of 70 seconds, gave a slightly better yield than the ML-100A material with a rating of 50 seconds. The latter gave a yield increase of 76% over the untreated check and a decrease of 38% in defoliation due to Septoria blight. When ML-100A was formulated with each of the five different fungicides used, the average yield increase over the check plots was 173%, with a decrease in defoliation by blight of 78%. The corresponding averages for the fungicides used without oil were an increase of 115% in yield and a 75% decrease in defoliation. In other words, the addition of the oil to the fungicides increased the yield over the same materials without oil by 26%, with an accompanying decrease in defoliation of only about 15%.

Tribasic showed the greatest response to the addition of oil with a decrease in defoliation of 16% and a yield increase of 48%. Ziram was second with a yield increase of 33%. Dyrene, which gave the largest yield of the fungicides when used alone, showed the least response to the addition of oil with a yield increase due to the addition of oil of only 8%. This variable effect of adding oil to different fungicides has been evident on other crops, particularly tomatoes, where any incompatibility between an oil and a fungicide seems to be accentuated.

An examination of the foliage in the differently treated plots in this experiment indicated quite clearly that not only were there somewhat fewer lesions due to infection by the causal organism of the disease, but also that there were many fewer pycnidia (the fruiting, or spore-producing bodies of the fungus) present in the dead areas of the leaves. This suggested that the oil was in some manner interfering with the sporulation of the fungus after infection had taken place. This possibility will be discussed further in connection with the data for 1959 and 1960.

In 1959 three spray oils and three different fungicides were among the treatments applied to celery at the Muck Crops Substation. The oils used were Phillips Base Oil No. 1 with a viscosity rating of 32 seconds, Mobilsol with a rating of 50 seconds, and the Sun Spray Oil No. 7 that was used in 1958. The three fungicides to which oil (Phillips Base Oil No. 1) was added were Phaltan, ziram, and Tribasic. The effect of the oils in checking defoliation due to late blight and in bringing about a yield increase was approximately the same in 1959 as in 1958. The average yield increase for the three oils over that of the untreated check was about 78%, with a decrease in defoliation of 40%.

The Sun Spray Oil with a viscosity rating of 70 seconds gave a considerably greater reduction in defoliation, and a greater increase in yield over the untreated check plots than did the Phillips Base Oil with a viscosity rating of only 32 seconds. This is similar to the experience on bananas where it is said that an oil with a viscosity below 65 to 70 SUS does not give control of Cercospora leaf spot, whereas oils with a viscosity rating of 70 seconds, or above, are much more likely to give good results. Unfortunately, from the standpoint of disease control, the less active Phillips Base Oil was chosen to use with the three fungicides. However, the addition of this oil to the fungicides did result in some average increase in yield and a slight decrease in defoliation when compared with the fungicides used alone. The fungicides used without oil increased the yield by 330% over the check plots, and when oil was added the increase amounted to 372%.

Counts of the pycnidia present in the Septoria lesions present on the foliage of the differently treated plots showed an average of 35 fruiting bodies per 30-x field (100 counted) of a stereoscopic microscope, whereas the corresponding averages for the oils alone, the fungicides alone, and the fungicides plus oil were 21, 25, and 24, respectively. The count was lowest of all treatments with Sun Spray Oil No. 7, where the average was only 12 pycnidia per 30-x field.

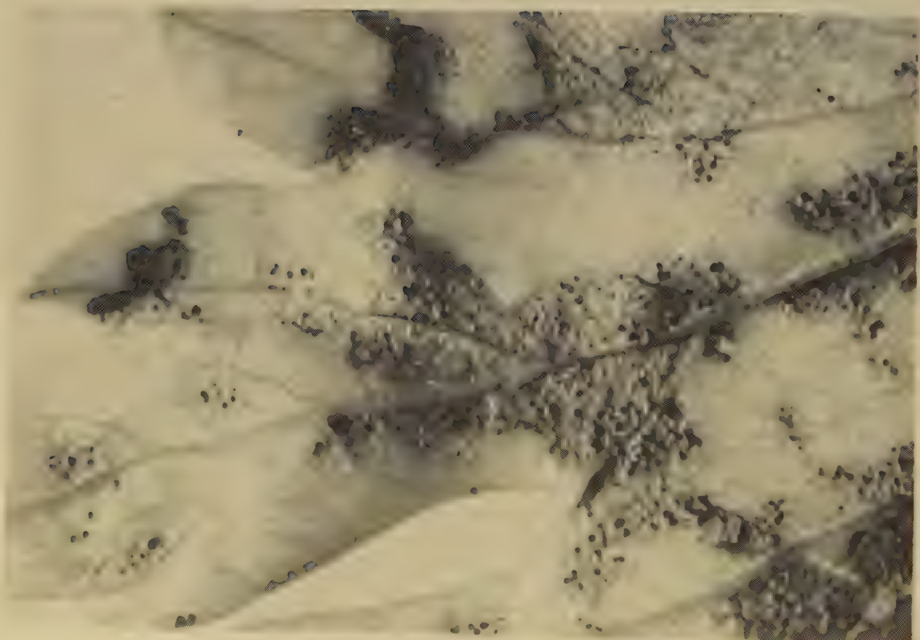

FIGURE 1. Pycnidia of Septoria apii on celery leaf from an unsprayed check plant.

FIGURE 2. A similar area on a leaf from a plant sprayed with oil.

Table 1. Influence of oils used alone or in combination with a fungicide on the control of Septoria leaf spot on celery in 1960.

Treatments	Rate of use/acre /application	*Yield in tons/acre of trimmed celery	% defoliation on Sept. 8	Septoria lesions/ leaflet	Number of pycnidia/ 30-x field
1. None (Check)	--	10.0	60	25.0	98.8
2. Phillips Base Oil #1	1.5 gal.	12.8	40	15.8	68.6
3. Sun Spray Oil #11	1.5 gal.	20.0	27	8.9	0.3
4. Tribasic	6 lb.	17.5	24	4.3	80.6
5. Tribasic + Sun Oil	6 lb. 1.5 gal.	22.8	10	8.3	0.1

*The LSD value at 19:1 was 1.0 ton/acre in a total of 16 treatments, most of which are not listed here.

Disease control and reduction of pycnidial (spore) formation in the Septoria lesions on the foliage sprayed with oil were more striking in 1960 than in either 1958 or 1959. In 1960 Phillips Base Oil No. 1 with its viscosity rating of 34 seconds was compared with Sun Spray Oil No. 11 with a rating of 118 seconds. The contrast between their performance in terms of disease control, yield, number of pycnidia formed, and the number of lesions per celery leaf was quite striking and confirmed still further the thesis that the more viscous oils are more effective in disease control than lighter ones. The data relative to the different criteria of performance are listed in Table 1.

Table 1 shows that the "heavier" oil gave by far the better disease control and much the greater decrease in the sporulation of the fungus. In fact, in this instance the oil (Sun Spray Oil No. 11) was more effective than Tribasic in checking pycnidial formation, although it was less able to prevent initial infection and the resulting defoliation of the plant than was the fungicide. When the two were used together sporulation was virtually eliminated and defoliation was well controlled, although for some reason initial infection, as represented by the number of lesions per leaflet, was greater than when Tribasic was used alone. The better control of defoliation resulted in a 30% increase in yield over Tribasic used alone.

The contrast in the appearance of lesions of Septoria apii on an unsprayed leaf and on one treated with Sun Spray Oil No. 11 is shown in Figures 1 and 2. There were at least 300 times as many pycnidia (the black dots visible in Fig. 1) per unit of lesion area on the foliage of the check plant as on the sprayed leaf; and although certain pycnidia started to form they ceased to develop before reaching their full size when oil was present.

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FLORAL INFECTION OF LADINO WHITE CLOVER, INCITED BY CURVULARIA TRIFOLII[1]

R. A. Kilpatrick[2]

Summary

Floral infection of Ladino white clover, incited by Curvularia trifolii, occurs in field plots each year at Dover, New Hampshire, and was artificially reproduced in the greenhouse. Flowers of all ages are susceptible. Symptoms consist of chlorosis of flower parts, wilting, and death of the petioles and flowers. Seeds fail to develop on severely infected flowers.

Curvularia trifolii (Kauff.) Boed., the causal agent of Curvularia leaf spot, is known to attack leaves and petioles of white clover, Trifolium repens (1, 2, 3). This note, however, appears to be the first published record of flower infection.

Naturally infected plants (Fig. 1A) have been found in the field each summer in New Hampshire since 1957. Flowers of all ages are attacked, turn chlorotic, wilt, and die. The fungus causes necrosis of the inflorescences, which gradually progresses into the petioles. Eventually the petioles die, forming a shepherd's crook. Seeds fail to develop on severely infected flowers. Isolation of Curvularia from field specimens is complicated by invasion of secondary fungi such as Alternaria sp. and Cladosporium sp., as well as others.

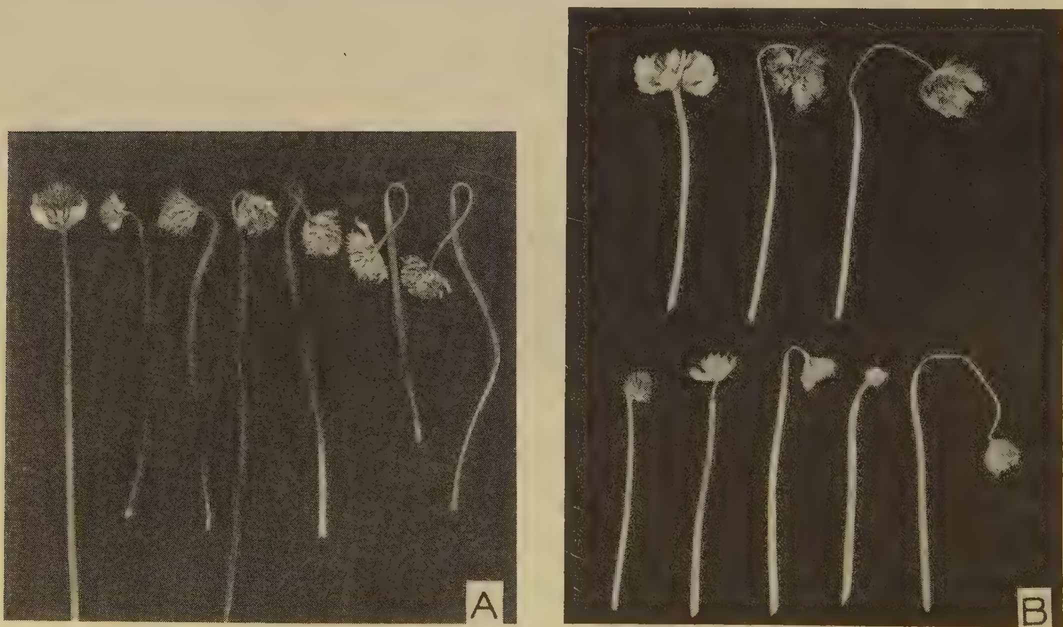

FIGURE 1. Flower symptoms on Ladino white clover, incited by Curvularia trifolii. A-- Field symptoms, natural infection. B-- Greenhouse symptoms, artificial infection. Left, healthy flower.

[1]Cooperative investigations of the Crops Research Division, Agricultural Research Service, United States Department of Agriculture and Departments of Botany and Agronomy, New Hampshire Agricultural Experiment Station, Durham. Published with the approval of the Director of the New Hampshire Agricultural Experiment Station as Scientific Contribution No. 267.

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Symptoms produced in the greenhouse were similar to those found in the field, but more severe (Fig. 1B). Flowering white clover plants were inoculated by the method used in an earlier study (3). Symptoms developed within 14 days and became progressively worse until flowers and petioles were completely killed. Leaf symptoms were lacking or few. The causal fungus was reisolated from infected inflorescences and petioles.

Literature Cited

1. BONAR, LEE. 1920. Wilt of white clover, due to Brachysporium trifolii. Phytopathology 10: 435-441.
2. KILPATRICK, R. A. 1958. Curvularia leaf blight of clovers and its causal agent, Curvularia trifolii. Phytopathology 48: 513-515.
3. LEHMAN, S. G. 1951. Curvularia leaf blight of Ladino clover. Plant Disease Reptr. 35: 79-80.

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STOLON DECAY OF COMMERCIAL SPECIES OF MINT IN INDIANA

Ralph J. Green, Jr.[1]

The propagation of peppermint (Mentha piperita) and spearmints (M. cardiaca and M. spicata) in the muck soils of northern Indiana is by stolons dug from the field in the early spring. These stolons develop slightly below the soil surface and are plowed down in the fall to reduce winter damage. In spite of this protection, there is frequently considerable mortality and loss of vigor of these vegetative organs. The volume and quality of stolons available for new plantings may, therefore, be sharply limited. In 1960 many growers attempted to increase acreages but, in many instances, were unable to do so because of a lack of planting stock.

Growers frequently attribute stolon deterioration to winter kill, and this undoubtedly is a contributing factor. Low temperatures during periods of low soil moisture or lack of snow cover may result in considerable damage to dormant stolons. However, another problem has been generally overlooked, namely the incidence of extensive stolon infection and decay by soil-borne fungi. Samplings of stolons during the past 2 years have revealed extensive infection and decay that appears to be a contributing factor to the decline and loss of vigor of stolons the following spring.

Stolons from several commercial plantings of peppermint and spearmint exhibited varying degrees of infection. The symptoms were similar, consisting of distinct, slightly sunken reddish-brown lesions with distinct borders (Fig. 1). The older lesions become soft and very extensive.

FIGURE 1. Symptoms of stolon decay on mint.

Laboratory isolations from the young lesions invariably yielded Rhizoctonia sp., while the older lesions were also invaded by Fusarium spp. and bacteria. Rhizoctonia spp. are common and persistent members of the microflora of the muck soils of Indiana and cause similar lesions on the non-fleshy portions of the stolons of potatoes grown in these soils.

This is the first report of the association of soilborne fungi with the decline and loss of vigor of mint stolons in commercial plantings in Indiana.

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RAPID IDENTIFICATION OF THE ONION PINK ROOT FUNGUS[1]

R. D. Watson[2]

Onion transplants, sets, and seed "mother" bulbs often are produced in one field or region of the country and then transplanted to other fields, farms or even regions for the onion production. If the pink root fungus is present in this propagating stock, it can be widely disseminated. Many new fields and farms annually become contaminated in this way. Pink root is not widely spread in Idaho at present, but it is increasing and the large commercial onion acreage is threatened by this destructive disease. A practical method of indexing transplants, sets and bulbs before planting would make possible the avoidance of spread of the fungus and save many farms from contamination.

Visual inspection of propagation plants is not always adequate for determining presence of pink root, as not all infected bulbs show distinct pink root symptoms. The dried roots of mature sets and bulbs lose the pink color and not all pink-colored roots contain the pink root fungus *Pyrenochaeta terrestris* (Hans.) Gorenz, J. C. Walker & Larson. Certain red pigment-producing forms of *Fusarium* will color onion roots. The isolation and identification procedures normally used by plant pathologists are too time consuming for general indexing for commercial production. In addition, the isolation procedures require considerable skill to separate the slow-growing pink root fungus from the rapid-growing and ever present *Fusarium* species. Some of the isolated cultures of *Pyrenochaeta* do not readily produce fruiting bodies which are necessary for positive identification of the fungus.

A quick method perfected for laboratory identification of *Pyrenochaeta* has proven valuable in preliminary screening of infected roots, and has speeded up the research program in the basic studies of the onion root rot disease complex. This method of indexing combined with bulb treatment could be of value to onion producers and seedsmen as a practical control for pink root.

METHOD

The pink root fungus *Pyrenochaeta terrestris* has been observed in the laboratory to produce red pigment when grown on wheat straw, a "weak" culture medium. The pigment may vary in color from pink to scarlet red and occasionally nearly black, depending upon the strain of *Pyrenochaeta*. Early in its development the pigment is pink to dark red, becoming black as its intensity increases. *Pyrenochaeta* is the only fungus known to produce a pink pigment on wheat straw; therefore, the presence of color indicates a positive test for this fungus. The pink pigment may be prevented or destroyed by fungi and bacteria growing in a rich medium that permits their abundant vegetative growth; therefore, a simple "weak" medium was used which permits a positive test whenever the pink root fungus is present, with only a rare interference from other microorganisms. The color does not often develop in less than 6 days and may take up to 21 days at room temperature.

To index onion bulbs for the presence of pink root fungus, place the surface disinfected roots or portions of the onion stem plate with roots attached on the test medium. The parasitic fungi grow out of the root and over the surface of the agar. The *Pyrenochaeta* turns pink on contact with the straw.

The roots were disinfected in one part household bleach to four parts water for 2 to 4 minutes. The culture medium was composed of 20 g Bacto-agar, 3 g sodium nitrate and 1 g magnesium sulfate in 1000 ml of water, sterilized in an autoclave at 15 psi for 20 minutes. Sterile agar was poured into sterile culture dishes and sterile chopped wheat straw was sprinkled on the surface of the agar before it set. The wheat straw was sterilized with propylene oxide gas. The chopped straw was placed in a fruit jar, 1 ml of propylene oxide liquid was added for each quart of capacity, and the jar was sealed tightly and allowed to remain for at least 24 hours before opening. (If the straw is dry it should be moistened slightly before the propylene oxide is added.)

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CONTROL OF ROOT-LESION NEMATODE, PRATYLENCHUS PENETRANS, ON NARCISSUS[1]

Walter J. Apt and Charles J. Gould[2]

Abstract

Soil fumigation is effective in controlling a nematode root rot of narcissus caused by Pratylenchus penetrans in western Washington. Nematode populations were reduced by soil fumigants containing dichloropropenes, chloropicrin, methyl bromide, ethylene dibromide, and dibromochloropropane in descending order of effectiveness. Fumigation increased yields of No. 1 King Alfred bulbs over non-fumigation. The value of fumigation has been substantiated commercially. High rates of methyl bromide and, to a lesser extent, chloropicrin and dichloropropene increased losses from Fusarium basal rot.

INTRODUCTION

The importance of nematode diseases of ornamental bulbous crops has been recognized for many years. Symptoms produced by the bulb and stem nematode, Ditylenchus dipsaci (Kuehn) Filipjev, on narcissus and those produced by the bud and leaf nematode, Aphelenchoides fragariae (Ritzema Bos) Christie, on Easter lilies are well known to bulb growers in the Pacific Northwest. A more recent development in this area is the nematode root rot of narcissus caused by Pratylenchus penetrans (Cobb) Filipjev & Stekhoven.

This root-rot disease was first observed in the Northwest by Hastings, et al. (5), who reported a narcissus disorder in British Columbia in 1932 and isolated a fungus, Cylindrocarpon radicicola Wr., and various nematodes, including Tylenchus pratensis de Man, from the short root stubs that remained attached to the bulbs of diseased plants. They concluded that Tylenchus was the primary cause of "root decline" and that the other nematodes and the fungus were secondary. Root-lesion nematodes were included in the genus Tylenchus until 1934 when they were placed in the genus Pratylenchus by Filipjev (2). In 1939 Hodson (6) concluded that "root eelworms" were the cause of the root-rot of narcissus in England. Jensen, et al. (9) recorded its occurrence in Oregon in 1951 and attributed it to Pratylenchus sp. Jensen (8) later identified the causal agent as P. penetrans. Gerretsen, et al. (3) reported a narcissus disease in The Netherlands which had been observed since 1917. According to Jensen, et al. (9) the symptoms of the disease reported in The Netherlands are very similar to those observed in Oregon. In 1956 Slootweg (10) reported that the primary cause of the root rot in The Netherlands was P. penetrans and that C. radicicola played a secondary role.

Root rot of narcissus is present also in the bulb-growing areas of Washington. Typical symptoms of the disorder have been observed in many narcissus fields in the Puyallup Valley, and root-lesion nematodes have been isolated from roots of diseased bulbs and the surrounding soil. The nematodes were identified as Pratylenchus penetrans.

The first evidence of nematode injury appears in the fall when the newly formed narcissus roots are attacked. The root tissues penetrated by root-lesion nematodes exhibit small necrotic lesions or dead spots, which are frequently reddish and later turn dark-brown to black. These lesions enlarge until considerable areas of root surface are rotted. Finally, the whole root system is decayed. In the spring, the aboveground symptoms of nematode root rot appear as distinct areas in the field. Examination of affected areas reveals that the foliage has yellowed, fallen, and withered prematurely. This early die-back occurs when the plants normally should be growing vigorously. Bulbs produced in infested areas do not increase in size properly and in some cases are no larger than the original planting stock.

The root-lesion nematode is very susceptible to desiccation (4) and was not recovered alive from dried roots of bulbs held in storage (7). It is unlikely, therefore, that planting stock would be responsible for the spread of the nematodes into new areas. Therefore, the chief concern with this plant pest is infested soil.

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[1]Cooperative investigations of the Crops Research Division, Agricultural Research Service, United States Department of Agriculture and Washington State University. Scientific Paper No. 2058, Washington Agricultural Experiment Stations, Pullman, Washington.

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Eliminating the nematode from infested fields is difficult because it can live on a wide variety of plants. Control by means of crop rotation becomes possible only when the alternate crop fails to maintain or build up a destructive population of nematodes in the soil. At present, no suitable rotation crop which is not a host to the root-lesion nematode in the Northwest is known. It has been reported in The Netherlands (10) that African marigolds grown on infested soil reduce the Pratylenchus population considerably, even though the marigold roots are heavily parasitized by P. penetrans. Such cultural practices as complete summer fallow, weed control, and roguing of volunteer plants help reduce the nematode population but they are not completely satisfactory. The most promising control measure at present appears to be soil fumigation.

This paper presents data comparing six nematocidal soil fumigants for the control of root-lesion nematodes and the resultant increased production of salable narcissus bulbs.

MATERIALS AND METHODS

In August 1956, five soil fumigants at various rates (Table 1) were applied as broadcast treatments to experimental plots in two different bulb fields heavily infested with root-lesion nematodes. One field was a typical bulb soil of Puyallup sandy loam, while the other was a Puyallup silt loam.

Prior to treatment the soil was worked to a seedbed condition and leveled. Soil temperature was above 68° F but did not exceed 72°, and soil moisture was comparable with that in a good seedbed. All liquid materials were applied at an 8-inch depth through a pressure-orifice system with a tractor-mounted chisel applicator. The soil was sealed with a cultipacker immediately after treatment. Methyl bromide was applied as a hot gas under a polyethylene tarpaulin. The tarpaulin was left in position for 48 hours.

Treatments were arranged in a randomized block of four replications. Plot size was 22 x 100 feet on the silt-loam field and 22 x 190 feet on the sandy-loam field except that the methyl bromide treatment plots were 22 x 60 feet.

The experimental plots were machine-planted with King Alfred narcissus planting stock 4 weeks after treatment. Seven rows of bulbs at 36-inch spacing were planted in each plot. These bulbs had been treated 2 to 4 hours at 110° F to eliminate bulb and stem nematodes and reduce the incidence of certain pathogenic fungi.

Composite soil samples were taken from each plot 30 and 90 days after treatments to determine the nematocidal efficiency of the test materials. Each composite sample consisted of 10 sub-samples taken along the three center rows of bulbs with a soil sampling tube. Aliquots of each thoroughly mixed sample were processed by the method of Christie and Perry (1), and the root-lesion nematodes were counted after an extraction period of 48 hours. All nematode collections were preserved in fixative and permanent glycerin mounts were made of representative species.

In July 1957, 50 feet of the center row from the middle section of each plot was harvested, and the bulbs cleaned, weighed, and graded. During this process, data were taken on the incidence of Fusarium basal rot, caused by Fusarium oxysporum f. narcissi Snyd. & Hans., in the harvested bulbs.

In August 1957, an additional experiment was established to test further the relative effectiveness of various soil fumigants (Table 2). Ethylene dibromide and two commercial formulations containing dichloropropene as the most active ingredient were applied in a sandy loam infested with root-lesion nematodes. Treatments were arranged in a randomized block of six replicates. Plot size was 17 1/2 by 50 feet. Five rows of hot-water treated King Alfred bulbs at 36-inch spacing were planted in each plot 3 weeks after treatment. The center yield row in each plot was planted by hand, with an exact number of bulbs of comparable size; the four border rows were machine-planted with commercial planting stock. Composite soil samples were taken and processed 30 and 90 days after treatment and the root-lesion nematodes were counted. In July 1958 the bulbs were harvested and data were taken on weights and grades.

RESULTS

The data from the 1956-57 experiment showing the effectiveness of the treatments in reducing the root-lesion nematode populations are presented in Table 1. The fumigants containing dichloropropenes (DCP), ethylene dibromide (EDB), chloropicrin, and methyl bromide had killed 98% or more of the nematodes at both locations 90 days after treatment. Dibromochloro-

Table 1. Influence of various soil fumigation treatments on the control of root-lesion nematodes, yield of King Alfred narcissus bulbs, and incidence of Fusarium basal rot of narcissus, 1956-57.

Soil fumigant and rate[a]	Number of root-lesion nematodes recovered following soil treatment[b]				% yield of No. 1 bulbs[d]		% Fusarium basal rot[d]	
	Field A[c]		Field B[c]		Field	Field	Field	Field
	30 days	90 days	30 days	90 days	A	B	A	B
Dichloropropenes (DCP):								
35 gallons/acre	2.5	2.0	2.5	5.5	40.6	5.2	1.1	2.2
45 gallons/acre	1.5	0.8	0.5	7.3	40.6	5.4	3.3	1.6
Ethylene dibromide mixture (EDB):								
9 gallons/acre	65.6	29.8	368.8	38.3	35.4	10.3	1.5	1.2
15 gallons/acre	5.3	6.8	210.0	9.0	32.6	0.9	0.6	0.5
Dibromochloropropane mixture (DBCP):								
9 gallons/acre	427.8	334.8	4224.0	123.3	30.0	3.9	1.2	1.1
15 gallons/acre	377.5	333.5	1774.0	67.3	29.7	7.7	2.1	0.6
Chloropicrin:								
25 gallons/acre	1.5	6.0	--	--	36.9	--	3.0	--
35 gallons/acre	1.0	2.8	1.8	0.0	38.9	6.1	4.9	0.8
50 gallons/acre	-- [e]	--	1.0	0.3	--	12.9	--	2.7
Methyl bromide mixture:								
1/2 lb/100 sq. ft.	40.5	6.5	--	--	33.5	--	1.9	--
1 lb/100 sq. ft.	7.5	1.8	--	--	36.7	--	4.8	--
2 lb/100 sq. ft.	0.0	0.0	--	--	37.7	--	7.7	--
None (control)	1852.3	1737.0	4001.0	3256.0	16.7	2.2	2.4	0.9

[a]Soil fumigants kindly supplied by Dow Chemical Company. The dichloropropenes were 1,3-dichloropropene and related chlorinated C_3 hydrocarbons. The ethylene dibromide mixture contained 83% ethylene dibromide by weight or 12 pounds/gallon. The dibromochloropropane mixture contained 25% by volume of 1,2 dibromo-3-chloropropane or 4.33 pounds/gallon. The methyl bromide mixture contained methyl bromide 98% and chloropicrin 2% by weight; and the chloropicrin was a technical grade.
[b]Figures given are average numbers of root-lesion nematodes recovered from 1 pint of soil from each of four replications.
[c]Field A is a Puyallup sandy loam; Field B is a Puyallup silt loam.
[d]% based on total number of bulbs of all grades harvested.
[e]Dashes indicate treatment was not made in that field.

propane (DBCP) mixture at 9 gallons/acre was 80.7% effective in the sandy loam (Field A) and 96.2% in the silt loam (Field B). The 15-gallon rate reduced the population to 80.7 and 98%, respectively, in Fields A and B. Control of the root-lesion nematode was reflected in both increased plant growth and yields of salable bulbs in 1957 (Table 1).

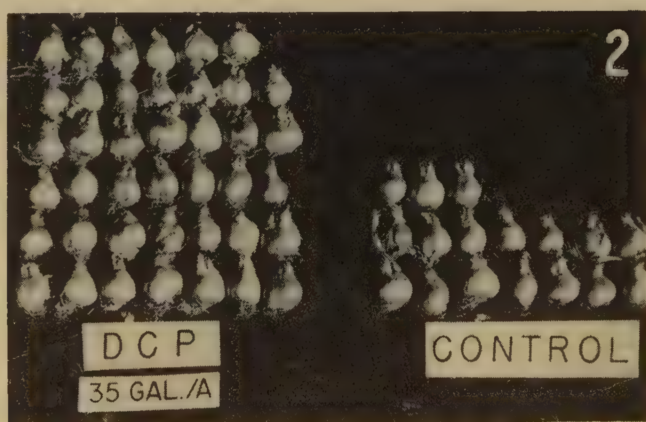
Aboveground symptoms of root-lesion nematode infections in the sandy loam became evident in the control plots in April and rapidly progressed during May and June. Plants growing in treated plots except those planted in the DBCP plots remained dark-green and produced vigorous growth throughout the growing season. In the DBCP plots, symptoms of infection began to appear in late May but progressed less rapidly than in the control plots. In the remaining treatments, plant growth continued through June when a natural maturation of the foliage began. Considerable differences were noticed between the root systems of plants grown in fumigated and non-fumigated soil. The plants grown in treated soil produced healthy vigorous systems of white roots. The roots of plants grown in untreated soil were short and rotted and often were absent (Fig. 1).

The differences in plant response to different treatments were particularly evident in the yield of top-grade DN-1 (double-nose) bulbs (Table 1, Fig. 2). The increased plant growth and resultant greater production of top-grade bulbs in treated areas were more evident in the sandy loam (Field A) than in the silt loam (Field B), but the differences in percentages of DN-1's in the control and variously treated plots were generally greater in the silt loam field. The

Table 2. Control of the root-lesion nematode by soil fumigation and yield of King Alfred narcissus bulbs, 1957-58.

Soil fumigant and rate (gallons/acre)	Number of root-lesion nematodes recovered following soil treatment[c]		% yield of No. 1 bulbs[d]
	30 days	90 days	
Dichloropropenes (DCP):[a]			
25	3.4	3.2	69.2
35	2.5	2.2	73.7
Dichloropropene-dichloropropane mixture (D-D):[b]			
25	2.6	2.2	74.6
35	1.3	0.9	77.4
Ethylene dibromide mixture (EDB):[a]			
5	94.6	28.2	67.0
10	55.4	12.2	69.4
None (control)	960.2	946.4	57.9

[a]See footnote to Table 1.
[b]Kindly supplied by Shell Chemical Co. Contained about equal parts 1,3 dichloropropene and 1,2 dichloropropane.
[c]Figures given are average numbers of root-lesion nematodes from 1 pint of soil from each of six replications.
[d]% based on total number of bulbs of all grades harvested.



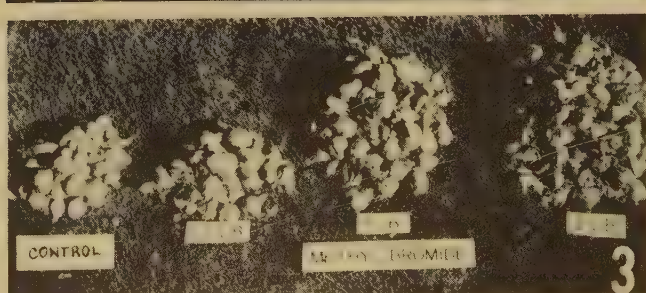


FIGURE 1. King Alfred narcissus bulbs grown in fumigated soil (left) and non-fumigated, root-lesion nematode infested soil (right).

FIGURE 2. Production of DN-1, King Alfred narcissus bulbs grown in a fumigated sandy loam (DCP, 35 gallons/acre) and non-fumigated, root-lesion nematode infested soil (control).

FIGURE 3. Increased incidence of Fusarium basal rot of King Alfred narcissus bulbs grown in non-fumigated soil (control) and methyl bromide fumigated soil (1/2, 1, and 2 pounds/100 square feet).

reasons for the overall low production of No. 1 bulbs in Field B are not exactly known, but a combination of other diseases and water-logged soil during the winter is believed to have contributed materially to it.

The incidence of Fusarium basal rot in the harvested bulbs was appreciably higher in certain of the treatments than in the controls (Table 1). This increase of the disease (Fig. 3) was especially noted in plots of the sandy loam treated with methyl bromide and to a lesser extent in those treated with chloropicrin and DCP.

The data from the 1957-58 experiment are presented in Table 2. Dichloropropene-dichloropropane (D-D) mixture, DCP, and EDB had reduced the root-lesion nematode population 96% or more by the end of 90 days. In contrast to the 1956-57 experiment, aboveground symptoms of the disease did not become evident in the control plots until the middle of June, shortly before harvest. In the treated plots, plant growth continued through June, when a natural maturation of the foliage began. Differences in production of No. 1 bulbs in control and treated plots were apparent (Table 2). Failure of the aboveground symptoms to develop fully was probably due to the very late spring and high moisture content of the soil throughout the growing season, which permitted continued growth with a reduced root system.

DISCUSSION

Slootweg (10) found that populations of 10 root-lesion nematodes per kilogram of soil resulted in serious root rot on narcissus. This apparent high susceptibility of narcissus to *Pratylenchus* is due partly to the fleshy nature of the roots and partly to the relatively small numbers of roots produced by the bulbs, which often are not replaced during the normal growing season.

Soil fumigants containing D-D, DCP, chloropicrin, EDB, and methyl bromide have relatively high vapor pressures and diffuse rapidly through the soil. The rapid buildup of these fumigants to a lethal concentration quickly reduced the nematode populations and protected the narcissus roots when they emerged from the bulbs in these tests. On the other hand, DBCP, with a low vapor pressure, probably diffused very slowly through the soil. Thus, at the end of 90 days the nematode population, although reduced, was still above the number necessary to produce considerable root rot. This lower degree of control was evident during the growing season and in the yield of top-grade bulbs. Control with DBCP could possibly be increased by earlier fumigation, if that could be worked into the cultural program of the grower.

The greater losses to Fusarium basal rot in methyl bromide and chloropicrin treated plots were rather unexpected. Most, if not all, stocks of King Alfred daffodils contain a few Fusarium-infected bulbs. The *Fusarium* is not controlled by the hot-water + formalin treatment. Therefore, the planting stock used in these tests presumably contained some diseased bulbs. However, observations indicate that the causal fungus does not usually spread from infested to healthy bulbs in western Washington soils. The lack of appreciable spread has been attributed previously to low soil temperatures during the growing season. However, the results with chloropicrin and methyl bromide indicate that presence of competitive organisms in the soil also may be a factor in retarding growth of the parasite.

The results of this investigation have shown that fumigation of soil with commercially available nematocides holds promise for the control of nematode root rot of narcissus in the Pacific Northwest. After the promising results in 1956-57, commercial trials were made by several bulb growers in the Puyallup Valley with fumigants containing DCP as the most active ingredient. Their results have corroborated the experimental results and such treatment has been adopted as a standard practice for infested fields.

Literature Cited

1. CHRISTIE, J. R., and V. G. PERRY. 1951. Removing nematodes from soil. Proc. Helminthol. Soc. Wash. D.C. 18: 106-108.
2. FILIPJEV, I. N. 1934. The classification of the free-living nematodes and their relation to the parasitic nematodes, Smithsonian Misc. Coll. 89: 1-63.
3. GERRETSEN, F. C., D. J. HISSINK, K. VOLKERSZ, and K. ZIJISTRA. 1927. Een onderzoek naar de oorzaken en de bestrijding van het z.g.n. van den wortel gaan van Narcissen en Hyacinthen. Versl. Landbouwkundige Onderzoekingen der Rijkslandbouwproefstations 32: 302-384. (English Summary Rev. Appl. Mycol. 10: 796. 1931.)

4. HASTINGS, R. J. 1939. The biology of the meadow nematode Pratylenchus pratensis (de Man) Filipjev 1936. Can. J. Research D. 17: 39-44.
5. HASTINGS, R. J., W. NEWTON, and G. STEINER. 1932. Root decline of narcissi. Plant Disease Reptr. 16: 112-113.
6. HODSON, W. E. H. 1939. Narcissus pests. Bull. Min. Agric. Fish. 51: 36.
7. JENSEN, HAROLD J. 1953. Observations on the root lesion nematode disease of narcissus and progress of control. Plant Disease Reptr. 37: 39-40.
8. JENSEN, HAROLD J. 1953. Experimental greenhouse host range studies of two root-lesion nematodes, Pratylenchus vulnus and Pratylenchus penetrans. Plant Disease Reptr. 37: 384-387.
9. JENSEN, HAROLD J., C. G. ANDERSON, and J. WIEMAN. 1951. A root-lesion nematode disease of narcissus. Plant Disease Reptr. 35: 522-523.
10. SLOOTWEG, A. F. G. 1956. Root-rot of bulbs caused by Pratylenchus and Hoplolaimus spp. Nematologica 1: 192-201.

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OPHIOBOLUS PATCH DISEASE OF TURF IN WESTERN WASHINGTON[1]

C. J. Gould, Roy L. Goss, and Maksis Eglitis[2]

Ophiobolus patch is a serious disease of turf in England and nearby continental areas, but only rarely has it been reported seriously affecting cultivated grasses in the United States. Since the turf disease complex in western Washington closely resembles that in England in other respects (1), it was suspected that sooner or later Ophiobolus patch would be found here.

Following a mild winter and very wet spring, typical symptoms of Ophiobolus patch appeared on a new experimental putting green turf area near Puyallup, Washington in June of 1960. However, perithecia of the causal fungus were not found until 5 months later, in November.

The disease appeared first as light brown spots of turf with diameters of only a few inches. However, most of the affected areas increased rapidly in size; some became 2 feet or more in diameter (Figs. 1 and 2). Both shoots and roots of the grass plants were severely attacked,

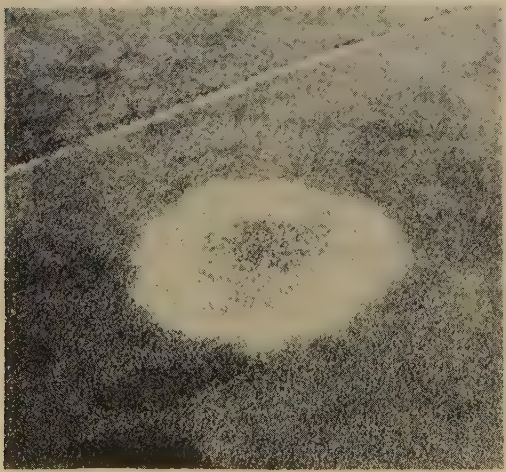

FIGURE 1. Typical appearance of Ophiobolus patch on Astoria Bent turf.

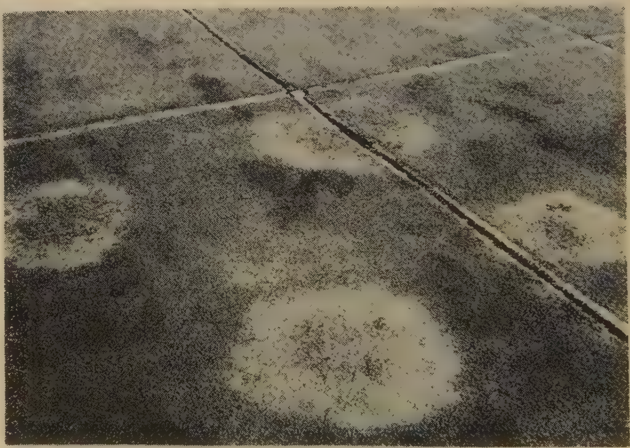

FIGURE 2. General appearance of an Ophiobolus infested area showing range of symptoms.

with the result that handfuls of dead turf could be pulled up easily. Affected areas did not recover for several months. The original species (Agrostis tenuis Sibth. hort. var. astoria) did not reinvade some spots and reinvaded others very slowly, starting in the center. Poa annua and various weeds became established in many spots so that the eventual appearance was that of miniature Fairy Rings. The disease was much more striking on the Agrostis putting green turf than on an adjacent lawn turf composed of 60% Creeping Red Fescue (Festuca rubra hort. var. Pennlawn) and 40% Astoria Bent. Fescue filled in the affected spots in the latter case. Smith (5) lists certain varieties of A. tenuis as susceptible and of F. rubra as resistant.

Similar spots appeared about the same time on Astoria Bent turf plots at Farm #1 at the Western Washington Experiment Station, 5 miles from the other site. An Ophiobolus with similar spore measurements was found recently in this material and also in bent grass collected in January 1961 from a fairway on a golf course near Tacoma, Washington.

The fungus more closely fits the description of Ophiobolus graminis Sacc. var. avenae E. M. Turner than that of the type variety (O. graminis var. graminis). Sizes of asci and of ascospores for the type variety are reported somewhat differently by various workers, but they are smaller than those for the variety avenae as shown in Table 1.

Ophiobolus graminis var. graminis causes the common "take-all" disease of cereals. It is worldwide in distribution and is reported from all areas of the United States except the extreme southeastern States (7). It is usually most serious in northern United States and in Canada. Sprague (7) states that it appears to be native to the Pacific Northwest.

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[1]Scientific paper No. 2078, Western Washington Experiment Station, Puyallup, Washington. Work was conducted under Project No. 1394.

[2]Plant Pathologist, Assistant Agronomist, and Research Associate, respectively, Washington State University, Western Washington Experiment Station, Puyallup.

Table 1. Lengths of asci and ascospores of Ophiobolus graminis var. graminis, O. graminis var. avenae and the Washington collection on Agrostis tenuis hort. var. astoria.

Fungus	Source of data	Asci	Ascospores
O. graminis var. graminis	Sprague (7)	90-115μ	60-90μ (mostly 70-80μ)
O. graminis var. avenae	Turner (8)	120-138μ[a]	80-140μ (mostly av. 101-117μ)
Wash. collection on A. tenuis	---	100-164μ[b] (av. 134μ)	88-124μ[b] (av. 100μ)

[a] Average range in length of three isolates.
[b] Average length of 50 asci and ascospores.

Because Ophiobolus is so widely distributed on Gramineae, it is surprising that it has been reported so seldom as a troublemaker on turf in the United States. In 1932 Monteith and Dahl (3) briefly mentioned its occasional occurrence. However, none of the recent general bulletins on turf diseases even lists the fungus. Some golf course superintendents from eastern Washington have reported seeing such diseased spots previously and Dr. Marion Harris[3] has stated that several years ago he found Ophiobolus in a sample of turf from a golf course at Walla Walla, Washington.

In his excellent description of this disease Smith (4, 6) stated that applications of lime favored development of the disease. Lime had not been used in our plots and the pH was 6.0.

These plots were located on an area that had been planted in 1957 to spring oats but the latter had not suffered appreciably from any disease according to the agronomist in charge. Cereals planted in the spring, however, are rarely attacked by Ophiobolus. The area had been intensively plowed, disked, and leveled in 1959 and a general fertilizer (14-14-14 at 300 pounds/acre) was applied prior to seeding on July 30. The area was designed for a combination fertilizer-irrigation test but treatments had not yet been initiated at the time the disease appeared.

Smith (4) reported that the disease could be controlled by use of ammonium sulfate or mono-ammonium phosphate fertilizer. He (4) and Jackson (2) also showed that certain organic mercury fungicides were beneficial. The fungus appeared to be suppressed in our plots following applications of PMAS (10% phenylmercury acetate) at 3/4 ounce in 10 gallons water/1000 square feet every 2 weeks.

Literature Cited

1. GOULD, CHARLES J. 1957. Turf diseases in western Washington in 1955 and 1956. Plant Disease Reptr. 41: 344-347.
2. JACKSON, NOEL. 1958. Ophiobolus patch disease fungicide trial, 1958. J. Sports Turf Research Inst. 9(34): 459-461.
3. MONTEITH, JOHN, Jr., and ARNOLD S. DAHL. 1932. Turf diseases and their control. Bull. U. S. Golf Assn. Green Sec. 12(4): 156-157.
4. SMITH, J. DREW. 1956. Fungi and turf diseases. J. Sports Turf Research Inst. 9(32): 180-202.
5. SMITH, J. DREW. 1958. The effect of species and varieties of grasses on turf diseases. J. Sports Turf Research Inst. 9(34): 462-466.
6. SMITH, J. DREW. 1959. Fungal diseases of turf grasses. Bull. Sports Turf Research Inst. pp. 57-61.
7. SPRAGUE, RODERICK. 1950. Diseases of cereals and grasses in North America. Ronald Press Co., New York. pp. 92-95.
8. TURNER, ELIZABETH M. 1940. Ophiobolus graminis Sacc. var. avenae var. n., as the cause of take all or whiteheads of oats in Wales. Trans. Brit. Mycol. Soc. Vol. 24, parts 3 and 4, pp. 269-281.

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FUNGI ASSOCIATED WITH WHITE CLOVER STOLONS IN SELECTED AREAS OF THE SOUTHEAST DURING MID-SUMMER, 1959[1]

James E. Halpin and States M. McCarter[2]

Abstract

Six locations in the Southeast were surveyed to determine the fungi associated with stolon rots of white clover during mid-summer. *Fusarium oxysporum*, *F. roseum*, and *Rhizoctonia solani* were found at all six locations and were generally more numerous than other species. *Colletotrichum destructivum*, *Curvularia trifolii*, and *Leptodiscus terrestris* were also present at all six locations, but were less numerous. Each of these species represented at least 10% of the isolates in one or more of the fields surveyed. The remainder of the fungi collected represented less than 10% of the total isolates from any one individual location with the exception of *Sclerotium bataticola* which, although collected at only three locations, represented 12% of the isolates from one of these.

In clover-grass pastures in the southeastern United States, white clover plants frequently die during the summer months. Thin stands result. Maintaining clover in southern pastures often depends upon volunteer plants. Survival of the resulting volunteer stand is uncertain because of competition from the grass, unfavorable weather, seedling diseases, or other hazards. White clover in this region grows more as a biennial or winter annual. Limited production results and the farmer fails to realize the maximum utilization for which the crop is intended. If it persisted as a long-lived perennial it would contribute much more to forage production. To attain this goal, the factors contributing to the loss of the clover must be fully understood.

Garren (1) reported the summer months as the period of maximum defoliation; that is, the period during which growth continues but defoliation exceeds growth. It is also a time of high temperatures with erratic precipitation. Stolon rots are usually associated with this summer problem and, because of the stoloniferous habit of growth of white clover, are thought to be of major importance in its failure to maintain rapid summer growth or even summer survival.

McGlohon (2) surveyed the fungi associated with white clover stolons during June and July 1958, in various areas of South Carolina and one adjacent area of North Carolina. He reported a consistently high percentage of *Fusarium* spp. in his isolates while species of other genera, such as *Rhizoctonia* and *Trichoderma*, although isolated from a high percentage of the stolons at certain times, were inconsistent in their prevalence. Other genera were isolated only in certain areas or infrequently throughout his survey.

The current survey was made to obtain more information concerning the fungi present in white clover stolons during a later period of the summer and within the time of maximum clover degeneration and stolon disappearance. At the same time, an effort was made to determine the species involved and to test these species for pathogenicity on white clover in pure culture.

Samples of diseased clover stolons were collected at three locations in South Carolina, two in Alabama, and one in Louisiana during the final week of July and the first 3 weeks of August 1959. All fields selected for sampling had been in the production of clover for 2 years or more and represented six different soil types (Table 1). Stolons were collected from live

Table 1. Location, date, and soil type of fields from which samples of stolon rots of white clover were collected, 1959.

Identification	Location	Date	Soil type
South Carolina No. 1	Upper Piedmont	28 July	Chewacla Sandy Loam
South Carolina No. 2	Upper Piedmont	26 July	Hiwassee Sandy Loam
South Carolina No. 3	Upper Piedmont	2 August	Eroded Cecil Clay
Alabama No. 1	Marion Junction	15 August	Houston Clay
Alabama No. 2	Tallassee	17 August	Cahaba Fine Sandy Loam
Louisiana	Hamburg	14 August	Delta Silt Loam

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[1] Technical Contribution No. 364 of the South Carolina Agricultural Experiment Station, Clemson.

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but unthrifty plants on which the presence of necrotic stolon lesions was observed. Only one stolon was selected per plant; however, several tissue samples from different parts of the stolon were plated out.

The collected stolons were brought into the laboratory, surface sterilized by dipping for 1 minute in a solution of 95% ethyl alcohol, then for 1 minute in a 1% solution of sodium hypochlorite, following which they were surface-dried within folds of sterilie absorbent paper. Tissue samples were made by removing narrow (1/8-inch wide or less) cross sections from regions of discoloration following the method described by Riker and Riker (3). Areas of extensive necrosis were avoided. Samples so obtained were then imbedded in potato-dextrose agar (PDA) in sterile Petri dishes and incubated at room temperature. Isolations were made from these plates at intervals of 3, 7, 12, and 16 days from the date of plating. The isolates were transferred to PDA slants and, after a suitable period for growth, stored in the refrigerator for later identification.

Certain species were present in all six locations (Table 2). These included *Fusarium oxysporum*, *F. roseum*, *Rhizoctonia solani*, *Colletotrichum destructivum*, *Curvularia trifolii*, and *Leptodiscus terrestris*. The first three of these species were found much more frequently than the last three in most of the locations. Each of these six species represented at least 10% of the isolates from one or more of the fields surveyed.

Table 2. Cultures of fungi isolated from diseased stolons of white clover at each of six locations in the Southeast during the period 26 July to 17 August 1959.

Fungus species isolated	South Carolina No. 1	South Carolina No. 2	South Carolina No. 3	Alabama No. 1	Alabama No. 2	Louisiana	Total
Fusarium oxysporum	66	68	72	48	69	38	361
Fusarium roseum	51	53	56	57	48	28	293
Rhizoctonia solani	22	37	33	22	27	14	155
Curvularia trifolii	11	4	11	55	10	7	98
Colletotrichum destructivum	12	3	2	5	26	35	83
Leptodiscus terrestris	3	30	7	13	12	17	82
Fusarium solani	13	19	17	7	8		64
Fusarium spp.	18		14		2	12	46
Colletotrichum trifolii	8			8	15	11	42
Sclerotium bataticola				1	13	24	38
Nigrospora			23	7	2		32
Trichoderma lignorum		7					7
Pythium spp.			1	2	2		5
Miscellaneous fungi	6	8	6	12		3	35
Total	210	229	242	237	234	189	1341

The remainder of the fungi collected represented less than 10% of the total isolates from any one individual location, with the exception of *Sclerotium bataticola*. It was obtained from stolons collected at only three of the locations, but represented over 12% of the isolates from one of these.

Literature Cited

1. GARREN, KENNETH H. 1955. Disease development and seasonal succession of pathogens of white clover. Part II -- Stolon diseases and the damage-growth cycle. Plant Disease Reptr. 39: 339-341.
2. McGLOHON, NORMAN E. 1959. Survey of fungi associated with white clover stolons. Plant Disease Reptr. 43: 22-24.
3. RIKER, A. J., and REGINA S. RIKER. 1936. Introduction to research on plant diseases. John S. Swift Co., Inc., St. Louis.

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LEAF SPOTTING OF ILEX CORNUTA FOLLOWING USE OF OVEX[1]

D. L. Gill[2]

Nurserymen and custom sprayers of home gardens frequently use O, O-diethyl O-p-nitrophenyl phosphorothioate (parathion) or S-(1,2-bis(ethoxycarbonyl)ethyl)O, O-dimethyl phosphorodithioate (malathion) alone, or combined with an ovacide, as a general insect clean-up spray. Several cases have been called to the writer's attention in which a leaf spot of the new foliage on *Ilex cornuta* and particularly on the cultivar Burfordii has followed such a spray. The spots are usually round, necrotic, and scattered over the leaves. In some cases a scorch developed. The appearance of these spots suggested a pathogenic origin, but no fungi or bacteria could be found associated with them either by microscopic examination or by plating bits of the affected tissue. Foliage developing after the initial spotting remained free of spots. In the cases observed where complete information could be obtained it was found that p-chlorophenyl p-chlorobenzenesulfonate (ovex) was used in the spray.

To determine whether the spotting was associated with the use of one of the insecticides mentioned, plants were sprayed April 29, 1960 with the following materials:

1. parathion 15% wettable powder (3 pounds to 100 gallons).
2. parathion as above plus ovex wettable powder 50% active (2 pounds to 100 gallons).
3. Ovotran (ovex) 50% (2 pounds to 100 gallons).
4. malathion 25% wettable powder (3 pounds to 100 gallons).
5. Unsprayed.

Two ounces/100 gallons of an emulsifiable polyethylene sticker-spreader was added to each spray. Three Burfordii plants and one cornuta plant 4 to 6 feet tall in a general landscape planting under high pines were sprayed with each material. Four smaller plants 2 to 3 feet tall, apparently cornuta seedlings, in a nursery row were also sprayed with each spray. These plants were under heavier shade.

On May 10 heavy spotting and scorching on the new leaves were present on the plants sprayed with the ovex alone or combined with parathion. It was not present on the unsprayed plants or on those sprayed with parathion or malathion alone. The spotting and scorch were more severe on the Burfordii plants than on the others. Less injury appeared to be produced where shade was heavier. The injury was more severe than that previously observed, apparently because the sticker-spreader used gave better coverage and sticking. Ordinarily sticker-spreaders are not used by nurserymen or custom sprayers. Foliage developing after the spraying did not develop the injury.

These tests and observations indicate that ovex or the diluent used is capable of producing a spot and scorch on *Ilex cornuta* and its cultivar Burfordii.

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[1] Cooperative investigations at Tifton, Georgia of the Crops Research Division, Agricultural Research Service, United States Department of Agriculture and the University of Georgia, College of Agriculture, Agricultural Experiment Stations. Published with the approval of the Director as Journal Series No. 80.

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EFFECTS OF SULFUR DIOXIDE ON THE REDUCTION OF POSTHARVEST DECAY OF LATHAM RED RASPBERRIES[1]

R. A. Cappellini, A. W. Stretch, and G. S. Walton

Summary

Postharvest decays of Latham red raspberries caused by Botrytis and Cladosporium spp. were effectively controlled with sulfur dioxide (SO_2). Berries were exposed to single and multiple doses of SO_2 for 20-minute periods at room temperatures (80° to 85° F) and subsequently stored at 50°, 70°, and 80° for 2 to 7 days. Initial fumigations were made approximately 4 hours after harvest, additional fumigations were either 24, 48, or 72 hours later. A softening and slight bleaching of the berries were noted when treated with approximately 0.50% SO_2. Although delayed effect on ripening was observed at approximate concentrations of 0.25% and 0.13%, it is not considered an injurious one. Off-flavors were not detected in the treated berries.

INTRODUCTION

The red raspberry is one of the choicest small fruits and also one which perishes most quickly after harvest. Decays resulting from both pre-and postharvest infections by Botrytis sp. and "overgrowth" by such fungi as Cladosporium and Alternaria greatly reduce the salable period and market value of the berries. Effective commercial postharvest methods to control the activities of these fungi are lacking. Results obtained during the past season with postharvest fumigations of SO_2 indicate the probability of using this gas as an efficient and economical method of prolonging the marketable quality of red raspberries. This appears to be the first report on the effective postharvest use of this gas on red raspberries.

EXPERIMENTATION

Field-run Latham red raspberries were exposed to single and multiple fumigations of SO_2 in a wooden chamber having a volume of 270 liters (Fig. 1). Various concentrations of SO_2

FIGURE 1. Equipment used in fumigating Latham red raspberries. From right to left; tank of SO_2, pressure regulating valve, blower, and flow-meter on top of chamber.

were obtained by delivering the gas into the chamber at 20 psi through a flow-meter. Initial fumigations were made 4 hours after harvest, additional fumigations were either 24, 48, or 72 hours later. All fumigations were for 20 minutes at room temperatures (80° to 85° F), during which the air-gas mixture was circulated throughout the chamber by means of an attached blower. Since analyses of the gas in the chamber were not made during the fumigation period, losses due to sorption and leakage were undetermined. The treated berries were stored at 50°, 70°, and 80°. Evaluations were made during a 2 to 7 day period after storage.

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Botrytis and Cladosporium spp. were the predominant fungi occurring on the berries in these tests. Cladosporium occurred chiefly in two and Botrytis in three of five tests completed.

Effects of SO_2 on Cladosporium: In preliminary tests, berries were fumigated with approximately 0.13%, 0.25%, and 0.50% SO_2. Four replications of 30 berries each were treated and then stored at 50° and 70° F; only the visibly molded berries were removed after each evaluation. Two 1-pint replications of berries were treated and stored at 80°, and all berries were discarded after each evaluation.

In a second test, single (0.25%) and double (0.25% + 0.13%) fumigations with SO_2 were made on two replications of 1 pint per evaluation. Storage temperatures were 50° and 70° F. All berries were discarded after each evaluation. The sound berries remaining after 7 days in the 50° storage tests were held for and evaluated after 2 additional days at 80°.

A highly significant reduction in decay was obtained in both tests (Tables 1 and 2). Treatment with SO_2 followed by storage at 50° F completely controlled Cladosporium for 4 days and permitted only slight activity of this organism after 7 days.

Table 1. The effects of various concentrations of SO_2 on the control of Cladosporium on Latham red raspberries.

% SO_2	% moldy berries for periods (days) of storage at three temperatures							
	50° F[a]			70° F[a]			80° F[b]	
	2	3	7	2	3	4	2	3
0	7	7	77	77	82	99	76	91
0.13	0	0	4	0	1	12	1	2
0.25	0	0	1	0	0	6	0	1
0.50[c]	0	0	0	0	0	2	0	0

[a]Average of four replications of 30 berries each.
[b]Average of two replications of 1 pint each.
[c]A slight bleaching and softening of the berries noted at this concentration.

Table 2. The effects of single and double SO_2 fumigations on the control of Cladosporium on Latham red raspberries.

% SO_2	% moldy berries for periods (days) of storage at three temperatures					
	50° F[a]		70° F[a]		80° F[b]	
	4	7	3	5	1	2
0	54	91	96	-	82	-
0.25	0	4	5	39	16	27
0.25 + 0.13	0	1	4	20	4	7

[a]Average of two replications of 1 pint each. Second fumigation made 24 hours after the first.
[b]Sound berries after 7 days at 50° F storage were removed to room temperatures for 2 additional days. Moldy berries removed after each evaluation.

Effects of SO_2 on Botrytis: A preliminary test was performed as outlined under the tests in which Cladosporium was the chief mold.

In a second test, single (0.25%) and double (0.25% + 0.13%) fumigations with SO_2 were made on four replications of 1 pint each per evaluation.

In a third test, five replications of 1 pint each per evaluation were exposed to single (0.25%), double (0.25% + 0.13%; 0.25% + 0.25%), and triple (0.25% + 0.13% + 0.13%) fumigations with SO_2. Storage temperatures in the second and third tests were 50° and 70° F; all berries were discarded after each evaluation.

Again a highly significant reduction in decay was obtained with all treatments at all storage temperatures (Tables 3 and 4). Excellent control of Botrytis was obtained with treatments followed by storage at 50° F for 4 days.

Table 3. The effects of various concentrations of SO_2 on the control of Botrytis on Latham red raspberries.

% SO_2	% moldy berries for periods (days) of storage at three temperatures							
	50° F[a]			70° F[a]			80° F[b]	
	4	3	7	2	3	4	2	3
0	15	17	63	47	90	98	39	97
0.13	6	9	27	9	22	29	8	26
0.25	0	3	15	2	7	21	6	25
0.50[c]	0	0	12	5	14	24	4	14

[a]Average of four replications of 30 berries each.
[b]Average of two replications of 1 pint each.
[c]A slight bleaching and softening of the berries noted at this concentration.

Table 4. The effects of single and multiple SO_2 fumigations on the control of Botrytis on Latham red raspberries.

% SO_2	% moldy berries for periods (days) of storage at two temperatures			
	50° F		70° F	
	4	7	3	5
0[a]	63	91	97	-
0.25[a]	7	29	27	75
0.25 + 0.13[a]	2	20	14	48
0.25 + 0.25[b]	3	16	3	45
0.25 + 0.13 + 0.13[c]	1	11	5	37

[a]Average from two tests with a total of nine replications of 1 pint each. Second fumigation made 24 hours after the first.
[b]Average of five replications of 1 pint each. Second fumigation made 48 and 72 hours after the first for 70° and 50° F storage, respectively.
[c]Average of five replications of 1 pint each. Second fumigation made 24 hours after the first: the third, 48 and 72 hours after the first for 70° and 50° F storage, respectively.

DISCUSSION

Sulfur dioxide, which has long been used effectively to prolong the storage life of grapes, may prove efficient and highly economical in prolonging the marketable quality of red raspberries. In tests performed during the 1960 season, two of the fungi most commonly associated with red raspberry spoilage after harvest were effectively controlled with concentrations of SO_2 tolerated by the variety of berries tested. Cladosporium, which initiates its activities after harvest, causing essentially an "overgrowth" on the berries, is more effectively controlled than Botrytis, which usually infects the berries before and continues its activities after harvest.

At an initial concentration of 0.50% SO_2, a slight bleaching and softening of the berries was noted, especially with those stored at room temperatures (80° to 85° F). However, some color deterioration was also noted in the untreated berries at this temperature. At initial concentrations of 0.25% and 0.13% SO_2 normal postharvest ripening of the berries appeared to be delayed but is not considered injurious. In fact, berries treated with these lower concentrations maintained a bright, freshly harvested appearance for several days after treatment.

Off-flavors were not detected in samples of berries consumed by the authors and various members of the department after treatment with SO_2.

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EPIDEMIOLOGY OF PEACH ROSETTE VIRUS IN PRUNUS ANGUSTIFOLIA

Glenn KenKnight

Abstract

Large numbers of chickasaw plum trees, Prunus angustifolia, were killed in Peach County, Georgia when peach rosette virus was experimentally introduced into thickets of that plum. Tests indicated that the virus spread through underground stolon roots and spread from top to top through the agency of an unknown vector.

Incidence of the phony disease of peach reached peaks in 1929 and in 1951 in Peach County, Georgia. At those times the disease appeared to threaten the local peach industry. Chickasaw plum, Prunus angustifolia, is commonly infected. This wild plum propagates by underground stolons, as well as by seed, and forms dense thickets in waste places. Peach growers, in cooperation with plant pest control agencies since about 1955, attempt to eradicate all plum trees within 300 yards of their orchards. However, the great numbers of wild plums more distant than 300 yards from peach orchards furnish seed for reinfestation of areas formerly cleared as well as inoculum for long-distance natural spread of phony peach virus.

Peach rosette (virus), for which the vector is unknown, strikes peach orchards at random throughout most, if not all, of Georgia but occurs in such low incidence that it is rarely detected in counties with low peachtree populations. Affected trees die. Occasional peach orchards, including a few in Peach County, have been devastated by the disease, but it has caused no extensive damage to commercial orchards in central Georgia for more than 20 years. Rosette is easily controlled in peach orchards by prompt roguing of affected peach trees and affected wild hosts at orchard edge. Every year for the past 12 years 1 to 3 affected trees in 3 to 7 peach orchards were noted in Peach County. During that period the disease occurred in at least 18 orchards, and the annual incidence was about 1 in 100, 000 peach trees.

During the same 12-year-period, peach rosette appeared to strike at random the wild plum thickets in Peach County. The disease killed 20 to 80% of trees more than 2 feet tall in affected thickets and often spread to neighboring thickets. Smaller plum trees usually escaped infection, and these, together with seedlings that emerged on the sites, eventually reestablished many devastated thickets.

For several years, beginning in 1949, peach rosette virus was artificially inoculated into several trees of widely separated plum thickets in Peach County. The epidemiology of the disease at each thicket location was observed.

METHOD

In May or June 1949-1953, buds or bark patches from peach or plum trees affected with rosette were grafted to trunks of small plum trees, one graft per tree near the ground line. At most locations five trees up to 5 yards apart in the center of a sizeable thicket were so inoculated. Thereafter the locations were visited several times each growing season to observe the initial appearance of rosette symptoms and the possible spread of the disease to neighboring plum trees.

RESULTS

After 2 years, successfully inoculated trees developed rosetted foliage, and a few to a great number of neighboring trees, presumably on a connected root system, ceased growth and became chlorotic. The inoculated and the chlorotic trees died, usually within 8 to 10 weeks after the initial appearance of symptoms. In some instances other plum trees up to 50 yards, but rarely farther, from the inoculated trees developed rosetted foliage at a later date as the result of natural spread of the disease. Trees inoculated by grafting or natural means usually developed conspicuously rosetted foliage, whereas trees infected through root connections usually did not.

Successful inoculations were made at nine well-separated locations. The disease spread naturally at five. The approximate percentages of trees killed by the virus and the approximate acreages involved in the locations where natural spread occurred were:

Location 1 - 70% of the old plums killed in 6 years in an area of 6 acres.
2 - 50% of the old plums killed in 9 years in an area of 1 acre.
3 - Active spread to single trees and small thickets but slight reduction in stand in 8 years in an area of 1 acre.
5 - 80% of stand, all large trees, killed in 5 years in an area of 1/2 acre.
9 - 100% of stand, all large trees, killed in 4 years in an area of 1/4 acre.

In the other locations the inoculated trees and some others died, but natural spread was not evident. The disease disappeared from all locations at the expiration of the time periods mentioned except on location 2, where peach rosette was still active in 1960.

DISCUSSION

Where single trees in a thicket of P. angustifolia were inoculated with peach rosette virus, all trees on a connected root system usually were destroyed. From the large reservoir of inoculum thus produced, natural spread to surrounding trees occurred in some locations. All trees were killed where thickets were old and more or less continuous, but spread was slower where thickets were young and discontinuous. Usually some old trees and most trees less than 2 feet tall escaped infection. Consequently the thickets were quickly reestablished.

Chemical control of wild plum trees, particularly in orchard environs, has been practiced in Peach County for a number of years. Usually some old trees are missed by the chemicals or survive the treatment, and seedlings soon emerge on the sites of treated thickets. Thus two or more follow-up treatments are required to complete the eradication job.

Where plum trees are large and the stand is dense, the cost of chemical control could be greatly reduced by first destroying a considerable part of the plum-tree population with rosette virus. However, biological control with rosette virus cannot be recommended as a supplement to chemical control, at least not until such time as the vector of rosette is determined and the vector's behavior in relation to dissemination of the disease can be studied. Occurrence of peach rosette in peach orchards often appeared to represent long-distance spread inasmuch as affected trees were as likely to occur in the center as at the edges of a 30- to 40-acre block of peach trees. The great increases in volume of inoculum that would result temporarily from a program of biological control of wild plum with rosette virus could be potentially hazardous to peach.

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FOMES ANNOSUS ON SLASH PINE IN THE SOUTHEAST

H. R. Powers, Jr. and John S. Boyce, Jr.[1]

Summary

Slash pine stands in three southeastern States were examined for signs of the root rot caused by Fomes annosus. This fungus was killing slash pines in 73% of the thinned plantations examined. Although the fungus occurred in a high proportion of thinned natural stands, there was less damage noticeable in these cases. Stands in which no cutting had been done, whether planted or natural, were almost free from damaging infections.

INTRODUCTION

Killing of slash pine (Pinus elliottii var. elliottii) by Fomes annosus was first reported in the Southeast in 1954 by Campbell and Hepting[2]. They recorded killing in a plantation in South Carolina and in a natural stand in Georgia. Both of these stands had been thinned. Since then, killing and windthrow due to annosus root rot have been identified in an increasing number of stands each year, primarily in thinned plantations. A survey was carried out during March 1960 to evaluate this situation. The overall objective was to obtain information on the occurrence and severity of annosus root rot in thinned and unthinned slash pine stands, both planted and natural. In each stand specific objectives were: 1) to determine whether the fungus occurred in the stand, and 2) if present, whether it was causing any damage.

METHODS

A total of 45 planted and 31 natural stands were examined in the survey. Since reports had indicated that disease losses were linked with thinning, it was decided that a ratio of three thinned stands for one unthinned stand would be examined. Moreover, limited observations had shown that slash pine mortality due to annosus root rot first appeared 2 to 3 years following a thinning. Wherever possible, therefore, stands that had been thinned 3 to 5 years previously were examined. To insure a representative geographic sample, stands were selected in four localities in South Carolina, four in Georgia, and three in Florida (Fig. 1). As far as possible, stands were located at random, with no prior knowledge as to the presence or absence of the disease.

An effort was made to examine as much of each stand as possible. Fruiting bodies of the fungus on stumps, dead snags, and living trees, and the characteristic white stringy rot of roots on windthrown trees were recorded. Occasionally diseased trees were located by symptoms such as yellowing and thinning of crowns. Each stand was first classified as having the fungus present or absent on the basis of signs mentioned above. In some cases, fruiting bodies were observed only on stumps with no apparent infection of living or dead trees. These stands were listed as having the fungus present but as having no apparent root rot damage.

Stands with evidence of infection in living or dead trees were classified as having either occasional or generalized infections. The "occasional infection" category included stands with one to several infection centers[3] which were somewhat localized. Many cases of incipient infection probably were not detected, and therefore classification of stands having "occasional infections" may have given a low estimate of the disease situation. "Generalized infection" included stands in which infection centers were numerous and generally distributed throughout the stand.

RESULTS

Signs of F. annosus were found in 82% of the thinned plantations and 69% of the thinned natural stands, but in only 11% and 8% of the unthinned plantations and natural stands, respectively.

Unthinned stands, both planted and natural, had little damage due to annosus root rot. Damage in thinned natural stands was common, with 52% of these stands rated as having "oc-

[1]Plant Pathologists, United States Department of Agriculture, Forest Service, Southeastern Forest Experiment Station, Asheville, North Carolina.

[2]Campbell, W. A., and G. H. Hepting. 1954. Fomes annosus on slash pine. Plant Disease Reptr. 38: 217.

[3]Infection center is an infected living or dead tree, or group of trees, where the fungus has worked outward from the initial point of infection. Usually, dead snags of annosus-killed trees are in the center of the area, with more recently attacked trees on the periphery.

Table 1. Occurrence of annosus root rot damage in four types of slash pine stands.

Type of stand	Number of stands examined	% stands in which lethal infections were: lacking	occasional	general
Plantations, thinned	34	27	47	26
Plantations, unthinned	11	91	9	0
Natural stands, thinned	23	44	52	4
Natural stands, unthinned	8	87	13	0

FIGURE 1. Location and number of slash pine stands examined in Fomes annosus survey.

casional infections" (Table 1). The most serious damage was in thinned plantations, with 73% of these stands having either "occasional" or "generalized" infections. Many of the most seriously affected thinned plantations had already sustained considerable volume losses.

DISCUSSION

Although F. annosus is considered indigenous in the southern United States, much of its damage may have gone unnoticed in the sawtimber economy of the past when natural stands were usually clear cut at ages greater than at present. Today's intensive pine plantation management, with its close, uniform spacing, root malformation and damage, thinning at early ages, frequent harvest cuts, and planting on drier, former agricultural sites, may be contributing to the greater incidence of annosus root rot in plantations.

Perhaps the most serious aspect of the annosus root rot situation is that the majority of the slash pine plantations in the Southeast have not yet reached thinning age. The survey results show that many of these plantations are likely to be invaded by F. annosus when they are thinned. The fact that 73% of the thinned plantations examined in the survey already had some losses due to this disease indicates that annosus root rot could become a serious management problem in planted slash pine in the Southeast. It is still too early to predict how general such damage will become, but reports from Europe, where this is the most serious forest tree disease, indicate that we are facing a new problem in our extensive stands of planted slash pine.

SOUTHEASTERN FOREST EXPERIMENT STATION, FOREST SERVICE,
UNITED STATES DEPARTMENT OF AGRICULTURE, ASHEVILLE, NORTH CAROLINA

RELATIVE SUSCEPTIBILITY OF FRAGARIA SPP. TO THE ROOT-KNOT NEMATODE, MELOIDOGYNE HAPLA CHITWOOD[1]

W. R. Orchard and M.C.J. van Adrichem

The alpine strawberry, Fragaria vesca L., and some varieties of the cultivated strawberry are known to be hosts for Meloidogyne hapla Chitwood, the northern root-knot nematode[2]. Varying degrees of susceptibility were found for 11 additional species and subspecies of Fragaria when the plants were grown in soil artificially infested with M. hapla.

Inoculum was prepared by introducing chopped roots of Chrysanthemum maximum var. Esther Read which were heavily infected with M. hapla into steam-sterilized soil in a greenhouse bench bed. This inoculum was increased by growing tomato plants var. Bonny Best in the inoculated soil for a period of 12 weeks. At the end of this period numerous egg masses of M. hapla were found on sample roots of lifted plants. At this time all tomato plants were topped at soil level and their roots were thoroughly mixed with the soil to provide a uniformly infested soil in the bed.

Eight young runner plants of each of the Fragaria species and subspecies were planted into the infested soil and allowed to grow for a period of 8 weeks (September 25 to November 25). At this time egg masses were found on roots of sample plants carefully lifted and washed. Table 1 shows the number of egg masses of individual plants counted with the aid of a stereoscopic microscope.

Table 1. Average number of egg masses per gram of root fresh weight.

Fragaria species	Plant accession number[a]	Average number of egg masses
Fragaria x ananassa Duch. nm. cuneifolia (Nut. ex Howell) St.	5116	112
F. chiloensis (L.) Duch. ssp. pacifica St.	5158	28
F. moschata Duch.		87
F. nipponica Mat.		30
F. orientalis Los.		182
F. vesca L. ssp. bracteata (Heller) St.	5102	123
F. vesca L. ssp. vesca		76
F. virginiana Duch. ssp. glauca (Wats) St.	5164	86
F. virginiana Duch. ssp. platypetala (Rydb.) St.	5157	0
F. virginiana Duch. ssp. virginiana	5159	25
F. viridis Duch.		7

[a]The nomenclature is that proposed by Staudt 1960. The numbers following certain entries refer to the identity of individual collections maintained with the Canada Department of Agriculture national reference collection of Fragaria species[3].

From the results, the resistance of F. virginiana ssp. platypetala is noteworthy. Some roots of this subspecies showed slight swelling but standard microscopic staining technique failed to show the presence of M. hapla. Plant response in terms of gall size was found to vary with different species and subspecies. The largest galls were found on F. virginiana and on Fragaria x ananassa nm. cuneifolia, the smallest on F. vesca ssp. vesca. Protrusion of the bodies of mature females from root tissue was more pronounced on the roots of F. virginiana ssp. virginiana than on those of other species. On F. moschata gall formation was weak and in some cases egg masses were observed without galling.

EXPERIMENTAL FARM, CANADA DEPARTMENT OF AGRICULTURE, SAANICHTON, B. C.

[1] Contribution No. 179 from the Experimental Farm, Research Branch, Canada Department of Agriculture, Saanichton, B. C.

[2] Anonymous. 1958. Host list for Meloidogyne hapla. U. S. Dept. Agr. Mimeo. 15 pp.

[3] Staudt, G. 1960. The Fragaria collection of the Plant Research Institute, Research Branch, Canada Department of Agriculture, Ottawa. Mimeo. 15 pp.

The authors are indebted to Dr. G. Staudt, Max Planck Institut, Cologne, Germany and to the Botanical Gardens, Moscow, U.S.S.R., for generously supplying seeds or plants.

DUTCH ELM DISEASE IN KANSAS IN 1960[1]

C. L. Kramer[2] and Hugh E. Thompson[3]

Dutch elm disease was first reported in Kansas City, Kansas in 1957 in both Wyandotte and Johnson counties. It spread into this area from Kansas City, Missouri, where it had appeared earlier. During the following season, 1958, the disease continued its local spread in the greater Kansas City area as well as moving north into Leavenworth County and south into Miami County.

In 1959 the disease was found in two additional areas -- one in Douglas, Franklin and Lyon counties, and the other in the southeastern corner of the State in Montgomery and Cherokee counties. At that time the isolated occurrence of the disease in that part of the State was not understood since it could not be located in three tiers of counties separating this area from the northern area of infection. However, in 1960 the disease was located in Joplin, Missouri, less than 20 miles east of Cherokee County. It is quite probable that the disease entered the State at two different points and is spreading as depicted in Figure 1.

In 1960 the disease had spread into several new locations in eight new counties. These include: one city in Atchison, one in Shawnee, one in Anderson, two in Linn, one in Allen, one in Bourbon, two in Neosho, and one in Crawford.

Collections were made by Hugh Thompson and LaVerne Calkins[4] while isolations were made by Robert Lichtwardt[5] and C. L. Kramer.

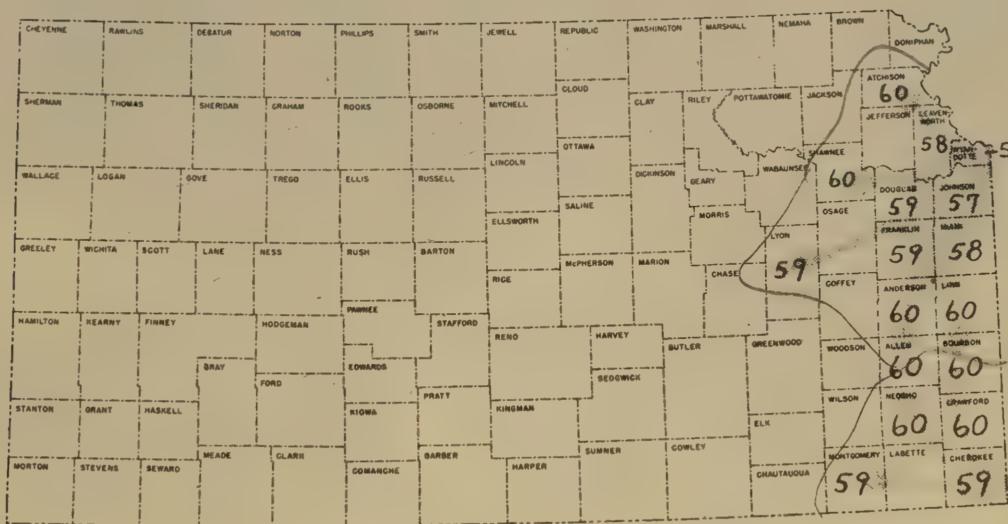

FIGURE 1. Numbers 57-60 indicate the years in which Dutch elm disease was first found in these counties, while the shaded areas depict the points where the disease seems to have entered the State and the directions it is spreading.

DEPARTMENTS OF BOTANY & PLANT PATHOLOGY, AND ENTOMOLOGY,
KANSAS STATE UNIVERSITY, MANHATTAN, KANSAS

[1]Contribution No. 574, Department of Botany and Plant Pathology, and No. 785, Department of Entomology, Kansas Agricultural Experiment Station, Manhattan. Botany serial No. 736.
[2]Department of Botany and Plant Pathology, Kansas State University, Manhattan.
[3]Department of Entomology, Kansas State University, Manhattan.
[4]Department of Entomology, University of Kansas, Lawrence.
[5]Department of Botany, University of Kansas, Lawrence.

A PORTABLE GAS SAMPLER SUITABLE FOR MEASURING ATMOSPHERIC OXIDANT

Gustave Silber[1]

The general problem of air pollution and the plant damage it causes in many instances is receiving greater recognition throughout the world. Elevated ozone concentration characterizes one of the more serious forms of air pollution[2]. In field and greenhouse studies of "weather fleck" of tobacco, caused by ozone toxicity, it was necessary to sample the atmosphere for its oxidant content. The primary parts and assembly of the apparatus found suitable are shown in Figure 1.

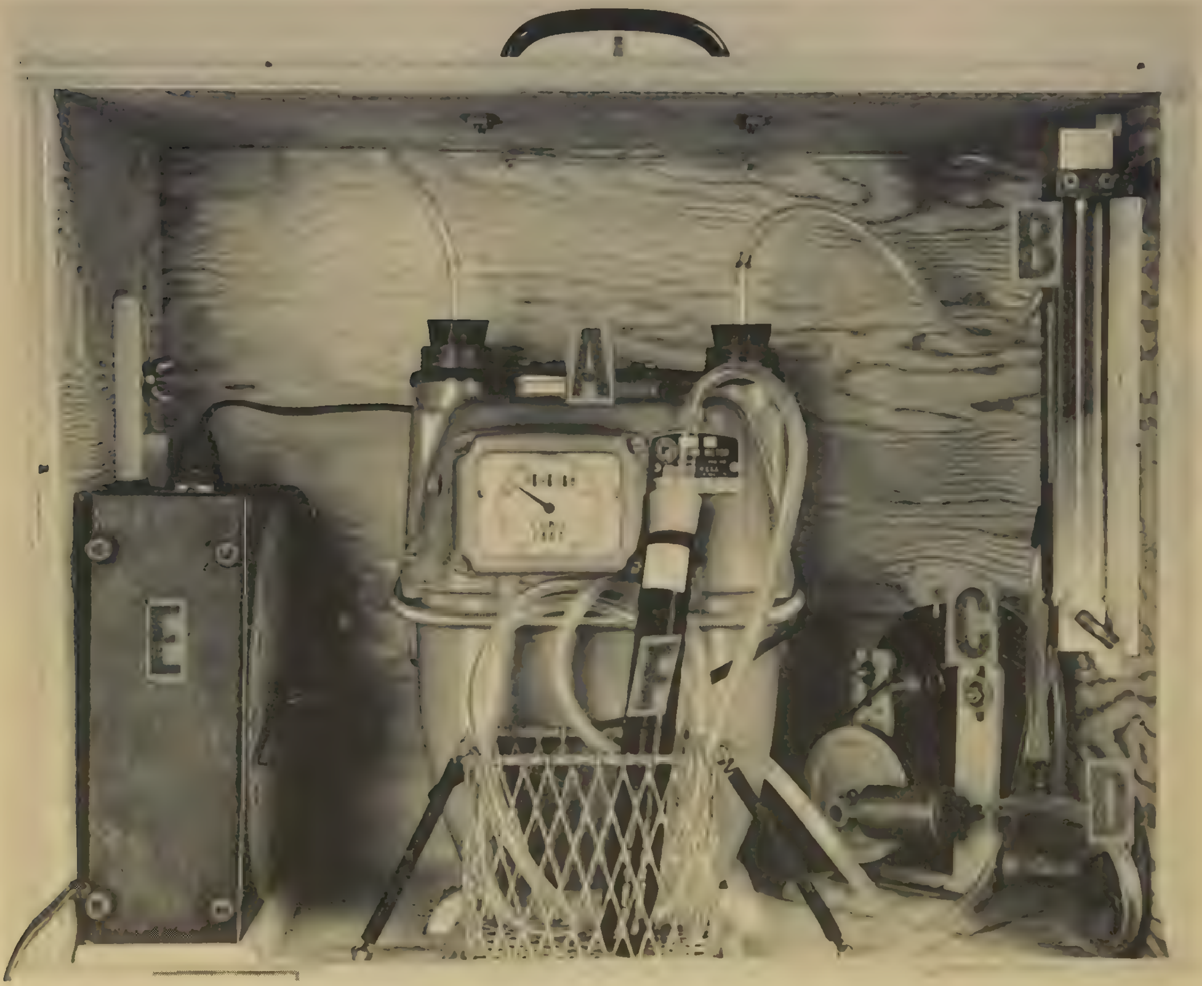

FIGURE 1. Primary parts of a portable gas sampler: A -- Gas meter. B -- Flowmeter. C -- Piston-type air pump. D -- Bypass valve. E -- Clock timer. F -- Gas scrubber.

[1]Plant Pathologist, Crops Research Division, Agricultural Research Service, United States Department of Agriculture.

[2]Heggestad, H. E., and J. T. Middleton. 1959. Ozone in high concentrations as cause of tobacco leaf injury. Science 129: 208-210.

A Rockwell[3] 150 gas meter (capable of metering 2.5 L. of gas per minute) measures the volume of air drawn through the collecting medium. This particular model gas meter has a dial marked to read 0.005 cubic foot, but 0.001 cubic foot can be estimated.

The rate of air sampling is measured by a Brooks[3] model 1355-V "Sho Rate" Rotameter (0.3 - 3.8 L. per minute). A modified Marvel Airflow[3] model C piston-type air pump draws the air sample. This type of pump, which is used ordinarily in a system in which gas is compressed, is easily converted by two simple adjustments to a system in which gas is evacuated. The pump is modified by 1) lowering the cylinder below the horizontal level and 2) reversing the ball check valve in the cylinder inlet line. These two adjustments cause the pump to exhaust air from the pump cylinder directly to the atmosphere.

The degree of vacuum at which the pump operates is varied with a bypass valve (Bunsen burner needle valve) installed in the line between the pump and the gas meter. If the bypass valve is omitted the efficiency of the pump can be adjusted by judicious positioning of the pump cylinder.

A clock timer can be set to shut off the pump after a fixed time interval has elapsed.

All the equipment is mounted in a plywood case for convenience in moving it from the laboratory to the field. Power requirements are sufficiently modest that the equipment can be operated in the field from an auto battery and a power converter.

For determinations of oxidants in the atmosphere the pump is set to draw gas through the collecting medium at a rate of 1.5 L. per minute by watching the flowmeter as the bypass valve is adjusted. Quantitative results are based on the volume of air sampled after temperature and pressure corrections.

Though the described equipment was assembled for determination of oxidant concentrations of the atmosphere, it will prove useful and economical for other investigations involving sampling of known quantities of gas for chemical or particulate content.

CROPS RESEARCH DIVISION, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, BELTSVILLE, MARYLAND

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[3]Use of name of a specific commercial product does not constitute endorsement by the United States Department of Agriculture.

AN ELECTRICAL AID TO PURE CULTURE ISOLATION

John M. Staley[1] and Howard Lyon[2]

An electrical microforge was devised for making soft-glass transfer needles used in single-spore and single-hypha isolation. The device (Fig. 1) consists of a 24-gage (B & S) chromel wire shorted across a two-prong male electrical plug mounted in a wooden stage that replaces the glass stage of a binocular dissecting microscope. Alternating current is supplied by a type 116 powerstat manufactured by the Superior Electrical Co., Bristol, Connecticut.

The voltage (0 to 5) and amperage (1 to 7 1/2) across the prongs are regulated so that the shorted wire glows with the desired intensity.

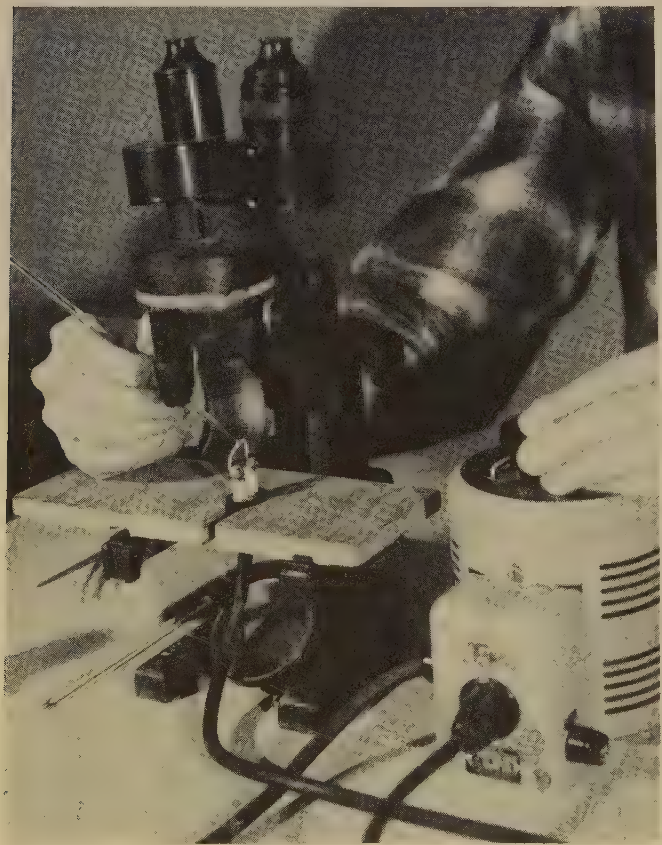

FIGURE 1. The electrical microforge, showing wooden stage, two-prong plug, glowing filament, powerstat, binocular microscope, and method of use.

To make the transfer tools, the end of a soft glass rod, first drawn to the desired size over a burner, is brought into contact with the glowing wire filament. On contact with the filament, the glass melts; and when pulled away it yields a thread or needle that is easily drawn to the desired size. The color of the filament is regulated by controlling the current, and the glass rod is manipulated to obtain the desired shape of tool. Precision of manipulation is obtained by observation through the binocular microscope.

Fine glass threads are made with a bright filament. Short, tapered needles can be made with a cherry-red filament. A dull red filament can be used to bend needles without breaking. The end of a fine glass needle held in close proximity to the glowing filament will form itself into a loop due to the unequal stresses caused by radiant heating. Such needles may also be beaded at the tip by passing through a burner flame. This makes a tool useful for picking up spores from flat agar surfaces.

[1]Plant Pathologist, Northeastern Forest Experiment Station, Upper Darby, Pa., and Graduate Student, Cornell University, Ithaca, N.Y.
[2]Visual Aid Technician, Cornell University, Ithaca, N.Y.

For complicated operations, tools can be reshaped quickly (and at the same time resterilized) for use in different ways. Since each tool is made and sterilized immediately before each operation, no special care is needed to protect tools in use or storage.

Transfer needles prepared with this microforge have been used to excise the agar surrounding a desired cell and leave the cell standing on an isolated pillar of agar. The cell can then be transferred by spearing and removing the agar pillar.

The forge, made from materials at hand, did not provide ideally sensitive control of the current passing through the wire filament. More sensitive control over the range of voltage and amperage required would be desirable. An air jet playing on the filament to aid in bending operations using radiant heat might also be desirable.

Though nichrome filaments of 24-26 gage burn out under constant use, they were inexpensive, easily replaced, and satisfactory for occasional use over a period of several days.

The only items required in addition to microscope and powerstat are a short piece of 2-strand electrical cord fitted with male plugs at each end, and the wooden stage. The prongs of one plug are tapped to accept brass bolts, which are used in fastening the 1.5-inch nichrome jumper wire. The wooden stage, grooved to fit between the clamps that normally hold the glass stage, can be made in any woodworking shop. Cost of these extra materials is estimated at less than two dollars.

The forge provides tools useful for manual work. Its advantages over more refined commercial models used in mechanical micromanipulation are lower cost and simplicity of use.

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UNITED STATES DEPARTMENT OF AGRICULTURE, UPPER DARBY, PENNSYLVANIA
AND DEPARTMENT OF PLANT PATHOLOGY, CORNELL UNIVERSITY,
ITHACA, NEW YORK

OCCURRENCE OF LATE BLIGHT DISEASE OF POTATOES IN MONTANA

M. M. Afanasiev

Late blight disease of potatoes, caused by Phytophthora infestans (Mont.) d By., was recorded for the first time in Montana in 1959 when a moderate outbreak of this disease occurred in potato plants of the Norland variety grown in an irrigated field near the city of Deer Lodge in Powell County. The seed of these potatoes was brought to Montana from another State. Other potatoes grown in the vicinity of the infested field and planted with Netted Gem variety were free of the disease. Tubers harvested from the field with affected plants were sold for table use and none were used for next year's planting. No new outbreaks of late blight disease of potatoes occurred either in the vicinity of Deer Lodge or in any other part of the State in 1960.

MONTANA AGRICULTURAL EXPERIMENT STATION, BOZEMAN

ANNOUNCEMENT

Dr. G. K. Parris, Chairman, Botany Department, Mississippi State University, State College, Mississippi has completed a compilation entitled "Index of Photographs in PHYTOPATHOLOGY, Vols. 1-41." This unbound, mimeographed manuscript of 233 pages, known as Miscellaneous Publication No. 2 of the Botany Department, Mississippi State University, and limited to about 130 copies, has been deposited with plant pathological libraries and also placed in the hands of a number of teachers of plant pathology in our country and abroad.

CORRECTION

REPORTER, March issue (Volume 45, Number 3). On page 229, in the last column to the right in Table 1, the temperature should be 5° C rather than 50° C. The Reporter regrets the error.